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PHYTOPATHOLOGY

An International Journal
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ABSTRACTS OF PAPERS ACCEPTED FOR PRESENTATION AT THE THIRTY-NINTH ANNUAL MEETING OF THE SOCIETY, CHICAGO, ILLINOIS DECEMBER 28 TO 30, 1947

The relation of pH to sporulation and growth of Gibberella zeae on agar and in liquid media. ANDERSEN, A. L. Sporulation and growth of *Gibberella zeae* are directly influenced by the initial pH of the culture medium, whether solid or liquid. The composition of the medium, particularly with respect to nutrient concentration, and nitrogen source, is also important, in part because of indirect effects on the pH changes that occur as growth progresses. On KNO_3-agar media, maximum sporulation resulted if the initial pH was approximately 3.0–3.3, whereas on NH_4NO_3-agar media an initial pH of 4.3 was optimum. In similar liquid media, maximum sporulation occurred in those media initially adjusted to pH 5–6. Nearly all the media shifted in pH as growth progressed. On KNO_3-agar media, this period of pH change was closely correlated with the period of rapid spore development; with those media having an initial pH below 7, the pH shift was toward alkalinity; those having an initial pH above 7 shifted first toward neutrality and then back toward the initial value. Media high in carbon and nitrogen produced more mycelium and fewer spores than did similar media low in these ingredients. Moreover, the optimum initial pH for mycelium production was higher for the low carbon and nitrogen media than in the case of media high in these two elements.

An evaluation of certain substituted phenyl esters for the treatment of cotton seed. ARNDT, C. H. Cotton seed naturally infested by *Colletotrichum gossypii* was treated with various substituted phenyl esters and the seed germinated in sand culture. At comparable dosages, the acetate ester of 2,4,5-trichlorophenol was effective in preventing seedling infection by *C. gossypii;* the formate and propionate were slightly less effective; while the carbonate, succinate, and laurate were ineffective. The substitution of bromine for chlorine caused little change in effectiveness. The nature of the chlorination of the phenyl acetate also influenced the fungicidal properties. The 2,3,4,6-tetrachlorophenyl acetate was more toxic to both the fungus and the host plant than the 2,4,5-trichlorophenyl acetate; while the chemicals with the Cl atoms in the 2,4,6, the 2,3,6, and the 2,3,4,5,6 positions were somewhat less effective. The latter was the most toxic of these chemicals to the host plant. The ortho, para, and the 2,4-dinitrophenyl acetates were not fully effective fungicides. Results obtained in field plantings, which were generally in agreement with the laboratory results, indicated that 2,4,5-trichlorophenyl acetate at a dosage of 1 gm./kg. of seed may be a satisfactory treatment for fuzzy cotton seed.

Inheritance of resistance to spot blotch of barley. ARNY, DEANE C. In seedling tests of F_2 and F_3 generations of several crosses, the difference between susceptibility and resistance to spot blotch (*Helminthosporium sativum*) appeared to be due to a single-factor pair, with susceptibility dominant. There was no detectable association of spot-blotch reaction with lemma and pericarp color (black vs. white), awn barbing or rachilla hair length, or reaction to stem rust. An association between spot-blotch resistance and the xantha character of Colsess IV was suggested in limited tests. In one cross spot-blotch susceptibility appeared to be associated with stripe (*Helminthosporium gramineum*) resistance.

Molds on apple storage containers and their control. ATKINS, J. G., JR. The growth of various molds on apple containers kept at high humidities is one of the problems associated with storage. *Penicillium expansum, Cladosporium herbarum, Stemphylium consortiale, Mucor plumbeus,* and *Cephalosporium* sp. were the most prevalent on new containers after several months in five western New York storage houses. Several other fungi were also encountered. *Mucor racemosus* was most frequently isolated from containers used in previous seasons on which the "whiskers" type of mold predominated. *Mucor plumbeus, Rhizopus nigricans,* and *Mucor* sp. were often present. Of the fungi commonly isolated from moldy containers only *Penicillium expansum* produced a rapid rot of Cortland apples. Other rot-producing organisms included *Botrytis cinerea, Aspergillus niger, Rhizopus nigricans,* and *Mucor* sp. A large number of chemicals were evaluated as mold inhibitors in laboratory tests. In such tests silver and mercury compounds were best followed by the Dowicides, Cuprinol, Thiosan, Copper 8-quinolinolate, and Spergonex. In storage tests Dowicides B, F, G and S, Thiosan, Spergonex, Cuprinol, Roccal, Phygon, and silver nitrate gave good control. Apples in direct contact with treated containers were injured by Spergonex, Cuprinol, and Dowicides B, F, G and S.

Physiological specialization in Cladosporium fulvum. BAILEY, D. L. The parasitic capabilities of the 8 physiologic races of *Cladosporium fulvum*, which have been reported to date in Ontario, Canada, are indicated in terms of reactions of the following differential host varieties: Red Currant (*Lycopersicon pimpinellifolium*); Vetomold; V–121; Stirling Castle; V–473; *L. hirsutum; L. hirsutum* var. *glabratum.* Evidence is offered that the evolution in virulence encountered in *C. fulvum* has been a function of the increasing resistance of the host varieties to which it has been exposed.

Time and locus of infection of American elms in relation to extent of vascular invasion by Ceratostomella ulmi. BANFIELD, W. M. Separate lots of American elm seedlings 18 to 28 feet in height were inoculated at intervals of one month, from April to September. Two holes per tree were cut to the cambium with a ¼-inch leather punch. The holes were packed with sawdust media containing *Ceratostomella ulmi* and then were taped. Less than 3 per cent of 318 inoculations made April 25 resulted in general vascular invasion. Fifty-three per cent of 146 inoculated May 10, 91 per cent of 103 inoculated June 7, and 70 per cent of 80 inoculated July 26 developed general vascular invasion and wilt. None of the 90 trees inoculated September 5 developed general vascular invasion. Similar inoculations made in apical shoots, 1- or 2-year stems, at comparable periods yielded various results. Inoculations made at any time in new shoots resulted in only limited invasion with inconspicuous wilt. Inoculations made in 1- or 2-year stems prior to May 10 or after September 1 resulted in only limited invasion in 850 trials. Five per cent of 199 inoculations made in June in 1-year stems and 17 per cent of 185 made in 2-year stems resulted in general invasion. Such invasion occurred in only 2 per cent of 167 inoculations made in July on 2-year stems and in none of 197 made then in 1-year stems.

Induced oospore production in the genus Phytophthora. BARRETT, J. T. It is well known that oospore production in certain species of *Phytophthora* is rare or erratic, or entirely unknown. In other species certain isolates give positive results, others negative. Pairing of different isolates of the same species and of different species by several students of the genus have resulted in stimulating oospore production. By the use of two isolates of *P. drechsleri*, one determined as male, the other female, and neither producing oospores alone, maleness and femaleness has been established in species which rarely or never produce oospores such as *P. citrophthora, P. cinnamomi,* and *P. infestans.* All isolates of *P. citrophthora* and *P. infestans* tested react as males, while those of *P. cinnamomi* react as females. All isolates of *P. cryptogea* from California react as males and one presumably from South Africa reacts as female. No oospores have developed as yet between *P. citrophthora,* female, and *P. cinnamomi* male. These results support, and extend in part, observations reported by Clinton, Leonian, Narasimhan, and Uppal.

Supplementary notations relative to spot anthracnoses (Elsinoë and Sphaceloma) in Europe since 1936. BITANCOURT, A. A., AND ANNA E. JENKINS. Examination of a culture and a leaf specimen discloses the presence in France of an hitherto unrecorded *Sphaceloma* on plum. We consider this most likely to be the same species as that found on plum in the Crimea in 1916 by Garbowski, who later reported it under the name of the related *Hadrotrichum populi* Sacc. Snowberry anthracnose, previously known only from North America, is here reported from France and Denmark. A revision of our 1936 tabulation of spot anthracnoses is offered with respect to present knowledge of their incidence in Europe.

Transmission of clover club-leaf virus through the egg of its insect vector. BLACK, L. M. In experiments started at the Rockefeller Institute, leafhoppers (*Agalliopsis novella*) carrying clover club-leaf virus (*Aureogenus clavifolium*) were allowed to deposit eggs in Grimm alfalfa plants (*Medicago sativa*). Later each of 30 nymphs was removed from the alfalfa as soon as it had hatched so that it had no opportunity to feed on the plant in which the egg was laid. Each nymph was placed on a crimson clover seedling (*Trifolium incarnatum*) and transferred at weekly intervals to a fresh seedling until it died. Three insects died within two weeks. The remaining twenty-seven lived from 13 to 33 weeks. The clover test plants and an equal number of control plants were grown in a greenhouse for at least six weeks after the removal of the insects. Twenty-four of the 27 insects or 89 per cent transmitted the disease. None of the insects infected plants until at least 3 weeks had elapsed from the time of hatching. Best transmission occurred during the 7th to 11th weeks inclusive. Altogether 68 of the 642 test plants and none of the control plants were infected. This is the second instance of a plant virus being transmitted through the egg of its insect vector.

The occurrence and transmission of Little Cherry in Washington. BLODGETT, E. C., E. L. REEVES, C. M. WRIGHT, AND H. E. WILLIAMS. The Little Cherry Disease recognized

in Washington first in 1946 in four orchards in three sections of the State was found to be widely scattered according to the 1947 survey. A total of 172,078 bearing cherry trees (about one-third of the State's total) was examined on 2,458 properties in 16 counties east, and in 5 west, of the Cascade Mts. A total of 289 infected properties was located in 13 counties east of the mountains, which represents a total of 1,471 trees affected or less than 1 per cent of those examined. It is believed that the disease has been in the State for about 5 years. Bud inoculation tests on June 27, August 19, and September 18, 1946 on six healthy Bing and two Lambert trees showed by mid-summer of 1947 that the virus had been definitely transmitted to four Bing and to the two Lambert trees. Of four root inoculated Bing trees, only one had the disease. Control of the disease is being attempted with a voluntary tree removal program.

Leaf rust of wheat in Mexico. BORLAUG, N. E., AND J. RUPERT. Leaf rust of wheat, caused by *Puccinia rubigovera tritici*, has become an increasingly important factor in wheat production in Mexico. Many varieties introduced into Mexico because of their known stem rust resistance have been found exceedingly susceptible to leaf rust under Mexican conditions. Furthermore, there is in Mexico at least one physiologic race of leaf rust not commonly found elsewhere and highly pathogenic to a wide range of varieties, including the durums. Mida and Trigo Supremo × 41–116 have been selected as the best parental material for leaf rust resistance. Newthatch and Kenya, commonly used as parents when stem rust resistance is sought, are highly susceptible to leaf rust races in Mexico.

Further studies on bacterial necrosis of the giant cactus. BOYLE, ALICE M. This paper summarizes the advances made in the study of bacterial necrosis of the giant cactus (*Carnegiea gigantea* (*Cereus giganteus*)) since the initial report before this Society at Philadelphia, 1940, and the publication of a paper on the disease in PHYTOPATHOLOGY in 1942. The disease is distributed over practically the entire habitat of the giant cactus; rapid spread is attributed to the activity of an insect vector now identified and proved culpable; longevity of the pathogen, age of the host with relation to susceptibility, and the astonishing attempts of the host to "cork-off" the bacterial lesions have been investigated. Serological studies support cultural, morphological, and other evidence that *Erwinia carnegieana* is an authentic species of bacterium.

Studies on plant teratomas. BRAUN, ARMIN C. When certain moderately virulent strains of the crown-gall bacterium were inoculated into *Kalanchoë* plants at different points along the stem, typical undifferentiated tumors, similar to those found on many other hosts, were produced. As the tumors enlarged those that originated in the younger, less differentiated portions of the stems produced leaves and stems that showed varying degrees of morphological differentiation. Many of these structures were highly distorted; others appeared to be more or less normal. Studies designed to determine whether these structural elements arose from normal host cells or from tumor cells strongly suggested that they were the result of the growth of tumor cells that had recovered in varying degrees from the effects of the tumor-inducing stimulus. The results can perhaps be interpreted by assuming the presence in plant cells of factors that determine the morphogenetic fate of the cells. In those instances, in which undifferentiated tumors developed, the factors concerned with morphogenetic determination must have been completely overwhelmed by the action of the tumor-inducing principle. The relative competence of the cells acted upon may therefore be of fundamental importance in determining the type of structure that ultimately develops.

Cytological effects of penicillin and streptomycin on crown gall. BROWN, J. G. Living cells in crown galls, as well as the concerned bacterial incitor (*Agrobacterium tumefaciens*), are killed by applications of penicillin and streptomycin to the galls; normal cells in and near the resulting necrotic galls remain visibly uninjured. Cytologic study reveals progressive stages in the destruction of the nuclei of the gall cells, including marked enlargement, sometimes to the degree of bursting of the nuclear membrane, followed by shrinkage and deformity; the nucleoplasmic network usually persists during the enlargement and into the stages of shrinking. That twigs, green leaves, and roots on aerial crown galls originate from normal cells, as maintained by Erwin F. Smith, rather than from totipotent gall cells, appears to be strikingly demonstrated in galls treated with the mentioned antibiotics: such organs remain alive for varying periods in galls otherwise necrotic from application of the antibiotics.

Anthracnose of cereals and grasses. BRUEHL, G. W. Anthracnose of Sudan grass and rye affects the major plant structures. In seedlings from infected seed the first internode is invaded, resulting in blighting or later crown infection and rot. Crown-

and culm-rot development diminished during the active vegetative growth of the plants and became active again as the plants matured. Yields were reduced by the crown-culm invasion. Spore production was usually heavy on the blighted plant parts of Sudan grass and less abundant on rye. Temperature response of isolates of *Colletotrichum graminicolum* indicated two groups; the one from *Sorghum* spp. more tolerant to higher temperatures than the cereal and grass isolates. Specialization on host groups was evident. Seedling blight of Sudan grass showed little temperature response between 16° and 32° C. Rye seedling blight and crown damage was reduced below 10° C. Preliminary evidence indicated an important antagonistic effect of soil microflora on *C. graminicolum*.

Aphanomyces cochlioides infestation in irrigated sugar beet–alfalfa rotation plots at Newell, South Dakota. BUCHHOLTZ, W. F. The sugar beet root-infecting fungus, *Aphanomyces cochlioides*, was abundant in plots devoted to a four-year rotation of sugar beets–oats and alfalfa–alfalfa–alfalfa. Plots in two similar four-year rotations, with potatoes or wheat in place of sugar beets, were apparently free of *A. cochlioides*. The continuous beet plot, without added manure or fertilizer, was infested, but not so heavily as those in the four-year alfalfa–sugar beet rotation. Two continuous beet plots, one with phosphate added and one with manure added annually, were apparently free of the organism. Plots planted continuously to alfalfa, potatoes, wheat, barley, oats, or corn were apparently free of *A. cochlioides*. Nonfertilized plots, planted to beets alternating with potatoes or oats, were infested. Plots in three-year sugar beet rotations without legumes were apparently free of the organism. In a six-year rotation of sugar beets–oats and alfalfa–alfalfa–alfalfa–alfalfa–potatoes, the third crop year alfalfa plot was heavily infested with *A. cochlioides*. A crop failure on this plot was successfully predicted in advance of field planting. Soil infestation by *A. cochlioides* apparently was "initiated" by crops of sugar beets, but very much favored by an abundance of alfalfa, a non-host crop, in the rotation.

Strawberry dud. CAMPBELL, LEO. The characteristic symptom of strawberry dud is a dearth of new root development, ranging from almost a full complement of new roots to none at all. Correlated with the root symptoms, an increased red discoloration and reduction in vigor of the tops to complete wilting and death of the plants follow transplanting. Poor stands and weak plants result. The tops of affected plants gradually acquire an open appearance and the younger, central leaves develop lighter green and are shorter than normal. Dud occurs most commonly on light, badly leached soils, innately deficient in nutrients. Faulty methods of handling plants, such as late digging, delayed planting, and poor or prolonged storage aggravate dud. Since the malady occurs where barnyard manure, green manure, or ordinary commercial fertilizers are used in abundance, some minor element deficiency is suspected as a causative factor. Such factors as overheating in the pack will result in symptoms of dud. Growing planting stock on good fertile soil and proper handling of plants after digging are being recommended as control measures, pending the determination of the primary cause of dud.

Effect of temperature on brown stem rot of soybeans. CHAMBERLAIN, D. W., AND W. B. ALLINGTON. The effect of temperature on the development of the brown stem rot of soybeans and on the causal organism was studied in the greenhouse and in the field. Air temperature appeared to be the important factor in disease development. When artificially inoculated plants were maintained at 15°, 21°, and 27° C., internal stem-browning appeared within three weeks in plants at 15° C., while plants at the higher temperatures showed little or no evidence of the disease. When these latter plants were transferred to the 15° C. chamber, they developed symptoms within 2 to 3 weeks. These results are correlated with outdoor temperatures in 1945, 1946, and 1947 and the development of brown stem rot in these years. In culture, the optimum temperature for growth of the fungus was 22–24° C.; for germination of conidia, 21–25° C.; for spore production, 15–20° C.

Relation between spore load on the seed and amount of smut infection in cereal grains. CHEREWICK, W. J. A method of determining smut spore load on cereal seed grain was developed in response to demands of farmers, and also with a view to conserve, during the war years, the short supply of seed disinfectants. This method was soon adopted by commercial firms that examine large numbers of farmers' seed samples and recommend whether or not seed treatment is needed. Greenhouse and field tests indicated that seed bearing spore loads of 1:1,000,000 or less by weight did not require seed treatment for smut control, and these results were used as the basis for the recommendations. Recent experiments revealed, however, that in the case of wheat seed threshed by a machine with rubberized cylinder and concaves, smut balls are frequently not crushed and, therefore, spores are not evenly distributed through the grain. While a sample of

such seed may show only a trace of spores it may develop up to 1 per cent smutted plants, when planted, owing to the presence of occasional smut balls. Moreover, in barley, false loose smut (*Ustilago nigra*) may be carried in part as mycelium under the hull. It would seem, therefore, that cereal seed carrying only a trace of smut spores must be treated for smut control.

Two important diseases of alfalfa in Manitoba. CHEREWICK, W. J. The most prevalent disease in Manitoba affecting both alfalfa and sweet clover is a crown rot which results in heavy losses, particularly in the alfalfa crop. Extensive surveys, isolation and inoculation tests during the past several years indicate that, under conditions prevailing in Manitoba, a strain of *Rhizoctonia solani* is the main, if not the sole, causal agent of the crown rot. The infected plants are readily invaded by saprophytic or weakly parasitic soil fungi which accelerate the disintegration of crown tissues and thus predispose the plants to winter-killing. The dead plants occur in patches which are soon overgrown with weeds and, therefore, damage occurs both from the reduction in forage yield and the infestation of the field with weeds. Another important disease is the "blossom drop" of alfalfa. It is prevalent in alfalfa seed growing districts in Manitoba and other provinces of Canada and frequently results in a complete failure of seed-set. The results of isolation and inoculation tests indicate that this disease may be caused by a species of *Alternaria*. The infected florets wither and fall off regardless of whether they were tripped or not. The addition to the soil of various essential food elements does not seem to have any effect on the disease.

The pathogenicity of Mycosphaerella citrullina. CHIU, W. F. Pycnospores, ascospores, and mycelium of *M. citrullina* were found equally infective on cotyledons and young foliage leaves of muskmelon and Hawkesbury and Improved Kleckley Sweet watermelons. Foliage leaves of cucumber, Hubbard squash, and Table Queen squash were very resistant. Infection of the hypocotyl and the stem of all cucurbits tested occurred only through punctures, through weakened areas, or by extension of lesions on the cotyledon. Cucumber and Hubbard squash were very resistant to formation of cankers on the hypocotyl when young, but both became very susceptible when old. The contrary was true for Table Queen squash. Cankers developed on the stems and hypocotyls of watermelon and muskmelon throughout their growing periods provided moist conditions prevailed. Ten isolates, secured by single-spore isolations and by irradiation, showed a gradation of pathogenicity from very high to none. The optimum temperature for infection of watermelon was 24° C., for muskmelon 16° to 20°. Field experiments revealed that Improved Kleckley Sweet watermelon was decidedly more resistant than Hawkesbury, while Cubit variety of cucumber was highly resistant compared with Stay Green, Chicago Pickling, Parker, Marketer, Long Stay Green, and National Pickling.

The effect of repeated passage of Helminthosporium sativum through the host on genetic variation and pathogenicity. CHRISTENSEN, J. J., AND C. L. SCHNEIDER. A line of *Helminthosporium sativum* derived from many successive monosporous isolations and grown on artificial nutrients for 28 years was passed through Marquis wheat ten successive generations. Disease-free seedlings were grown in test tubes containing water agar and were inoculated with single conidia of the pathogen. Monosporous cultures were made by atomizing the surface of potato-dextrose agar in Petri plates with a dilute suspension of spores from diseased plants after each generation in the host. These cultures were compared with isolates grown continuously on potato-dextrose agar. With the exception of five mutants, 14,400 monosporous progenies from the host were identical in cultural characters with the original line. The frequency of mutation of *H. sativum* on the host was approximately 1:2900. Cultures of the five mutants derived from the host were strikingly different from each other and their parent in color, type and rate of growth, and in amount of sporulation. Pathogenicity tests were made with monosporous isolates of the original line, obtained after five generations on the host, by sowing the seed of wheat and barley in infested soil. The virulence of the line was not changed in five successive passages through wheat plants.

Breeding tobacco for wildfire resistance. CLAYTON, E. E. Resistance to tobacco wildfire was obtained from *Nicotiana longiflora*, a small wild species. A fertile tobacco-like genotype, TL 106, was secured from the first backcross (Nt × Nl × Nt). TL 106 was crossed with various susceptible commercial tobacco varieties and F_2 seed grown. F_1 plants were also backcrossed to the same susceptible varieties. The F_2 populations gave 1,219 susceptible and 232 resistant plants, a ratio of 5.25 to 1. The first backcross populations gave 1,490 susceptible and 36 resistant, a ratio of 41.4 to 1. Tests with more advanced backcrosses showed that there was much variation in the number of resistant plants recovered from populations with identical parentage. The results indicate that

considerable chromosome irregularity still persists as the consequence of the interspecies cross, but suggest that as this is eliminated segregation on a simple basis is likely. The resistance was maintained even in the presence of prolonged, storm induced, leaf water-soaking. Field studies of resistant progeny from third and fourth backcrosses to susceptible tobacco showed that resistance was not linked with any undesirable growth characters.

The use of potato starch in virus purification. COCHRAN, G. W. Active tobacco-mosaic virus was purified by several procedures involving passage through starch columns. The virus was concentrated at the top of the columns by a "salting out" with ammonium sulphate or by a precipitation at the isoelectric point. In either case the passage of a pH 7, 0.1 M phosphate buffer through the starch moved the virus downward so that it could be collected at the bottom of the column as a purified concentrate. A more rapid method using extremely short starch columns was devised to fractionate the ammonium sulphate precipitable constituents of diseased plant juices. The ammonium sulphate concentration, just below the threshold of virus precipitation, was determined. The virus-protein mixture was adjusted to this concentration and passed through a thin starch filter. The salt concentration was then raised to a point slightly above the threshold of precipitation and the virus-protein mixture was passed through a second thin starch filter. The virus was held on the top of this second filter and was washed down with unsalted buffer and obtained as a purified concentrate.

Development of fungicidal aerosols as foliage protectants. COHEN, SYLVAN I. Thirty-four chemicals representing thirteen organic classes were tested for solubility in or compatibility with freon-12, then screened as an aerosol solution for phytotoxicity and fungitoxicity. Promising fungicides were formulated as treatments for the control of five foliage diseases. One aerosol treatment using an organic copper formula produced 96.5–100 per cent protection for 15–30 days against rose black spot, bean powdery mildew, bean anthracnose, and tomato early blight. It also succeeded in eradicating the pathogen from 24-hour-old lesions of bean powdery mildew. Two treatments at 15- and 22-day intervals reduced rust 39.1 to 60.6 per cent on three carnation varieties. A method was found to increase the deposit of aerosol droplets upon under and vertical surfaces sufficient to inhibit the spore germination of two fungi. Certain aerosol deposits exhibited good tenacity, as evidenced by no appreciable loss in fungitoxicity in laboratory tests after 4 minutes of washing. Aging of promising aerosol deposits for 17–30 days produced little or no effect upon their fungitoxic properties. The efficiency of aerosol deposits is indicated by the observation that when only 0.23 per cent of the total surface area was occupied by aerosol droplets of one formula, 100 per cent inhibition of *Stemphylium sarcinaeforme* spore germination resulted.

Forecasting late blight in eastern Virginia in 1947. COOK, HAROLD T. The method of forecasting late-blight epiphytotics described last year was put to a practical test during the 1947 season. Beginning on May 8, the cumulative rainfall and mean temperatures were plotted each day. Forecasts based on the graphs were issued at weekly intervals from May 15 to June 5. All of these forecasts stated that the weather conditions were unfavorable for late blight and that spraying or dusting to control the disease was not necessary at that time. The forecasts proved to be correct. There was no epiphytotic in Eastern Virginia and over 90 per cent of the potato and tomato acreage was entirely free of the disease. The crops also were free of other serious leaf and fruit diseases. Since it would have cost approximately $2,000,000 for routine spraying or dusting of the 60,000 acres of potatoes and tomatoes, the elimination of the needless use of fungicides is of considerable economic importance. A graph showing the moving 7-day average rainfall and temperature was developed to supplement the cumulative graph. The moving graph was more accurate for analyzing the late blight—weather relations over the entire season and also aided in correcting for unusually heavy rainfall early in the forecasting period.

Antibiosis and sugarcane root rot. COOPER, W. E., AND S. J. P. CHILTON. During the past two years, more than 6500 isolates of *Actinomyces* were obtained from Louisiana sugarcane soils. These were tested in culture to determine which ones were antibiotic to *Pythium arrhenomanes*, one of the important fungi causing root rot of sugarcane. The four major soils showed a consistent difference in the number of antibiotic *Actinomyces*. The Yahola silt loams had the highest number, the Yazoo silt loams next, the Iberia silt loams and first terrace soils were third, and the Sharkey clays the lowest. Comparative yield tests during 5 years with sugarcane on the various soils gave yields correlating with the number of antibiotic *Actinomyces*. In greenhouse tests in sterile soil, certain of the *Actinomyces* increased in the soil and reduced root rot in corn. Other strains failed to increase in the soil.

Stem anthracnose of Lima bean and its control. COX, ROBERT S. Stem anthracnose (*Colletotrichum truncatum*) causes serious losses of Lima beans in North Carolina. Although the most striking symptoms are brick-red lesions on pods, stems, and leaves, lesions commonly occur on the inflorescence, frequently resulting in blossom-drop and death of immature pods. The spore mass is typically gray, but a pink one is occasionally produced. In culture this pinkness is retained indefinitely. Experimental evidence indicates that the fungus is seed-transmitted. Of 16 varieties tested, all were highly susceptible, except Jackson Wonder, which showed some tolerance. Fungicidal sprays and dusts were applied at weekly intervals on replicated plots of the Henderson Bush variety. Phygon spray gave most effective control on foliage, stems and pods. In the spray test, average yields of marketable pods (lbs. per acre) from Phygon (1.5–100), Fermate (3–100), and control plots were 5847, 4803, and 1711, respectively. Of the dusts tested (Phygon, 5 per cent; Dithane Z–78, 6 per cent; Fermate, 10 per cent; Zerlate, 10 per cent; and tribasic copper sulphate, 5 per cent metallic copper), only Phygon and Dithane significantly controlled the disease. Phygon (dust or spray) caused slight leaf chlorosis and blossom-drop. Tribasic copper caused moderate foliage injury.

The effect of temperature and nitrogen source on the development of stem rust of wheat. DALY, J. M. The effect in sand culture of 40 three-salt nutrient solutions on the reactions of three spring wheats to race 56 of *Puccinia graminis tritici* was studied at temperatures above 80° F. At these temperatures, mineral nutrition did not alter stem rust reaction, although nitrogen concentration ranged from 20 to 305 p.p.m. Mindum was resistant (infection types 0; to 1); Marquis was susceptible (infection types 3 to 4) and Thatcher was susceptible (infection types 3 to 4) except in one experiment during a period of low light intensity when Thatcher was resistant (infection types 0; to 1). At temperatures below 70° F., certain forms of nitrogen affected the stem rust reaction of Thatcher, but not Mindum, wheat. Thatcher was mesothetic (infection type X) when grown in a mixture of three parts sand and two parts loam and supplied with 400 p.p.m. of nitrogen as $Ca(NO_3)_2$ or KNO_3; but plants supplied equivalent nitrogen as $(NH_4)_2SO_4$ were resistant (infection types 0; to 1). Applications of NH_4NO_3 resulted in resistance or moderate resistance to stem rust. The pH of the nutrient solution did not change the effect of nitrogen. Applications of $Ca(NO_3)_2$, $(NH_4)_2SO_4$, NH_4NO_3, or urea did not influence either the infection type or percentage of stem rust in field plots.

Inhibition of tobacco streak virus by juice from healthy plants. DIACHUN, STEPHEN. Usually numerous local lesions develop on carborundum-dusted tobacco leaves rubbed with inoculum prepared by crushing streak-infected leaves of *Nicotiana rustica* with 0.1 M Na_2HPO_4, but at times the number of such lesions is small. Thorough crushing of leaves in preparation of inoculum is one factor that reduces the number of local lesions, as compared with inoculum not so thoroughly crushed. Addition of juice from healthy leaves of *N. rustica* or *N. tabacum* to inoculum prepared from infected *N. rustica* also reduces the number of local lesions produced by the inoculum. It is possible that such an inhibitor may interfere in studies on concentration or rate of multiplication of virus in infected plants. Tobacco mosaic virus is not inhibited by juice from healthy tobacco plants.

Aspects of fungicidal control of snapdragon rust. DIMOCK, A. W., AND K. F. BAKER. Injury to snapdragons (*Antirrhinum majus*) from rust (*Puccinia antirrhini*) results from (1) a comparatively benign host-parasite interaction, (2) desiccation of invaded areas, (3) invasion by secondary organisms through rust-infected tissues, the type of injury depending on moisture conditions. With high inoculum potential the rust fungus was not controlled by weekly applications of Bordeaux mixture (8–8–100 plus DuPont Spreader-Sticker 1–1000) under either moist conditions at Ithaca, New York, or semi-arid conditions at Los Angeles, California. At Ithaca, nonsprayed snapdragons were only slightly damaged by desiccation, but all leaves were killed by secondary organisms. Bordeaux, though failing to control rust, gave almost complete control of secondary organisms and thus prevented loss of foliage, so that the plants appeared vigorous. Under dry conditions principal injury is from desiccation, and secondary organisms are not involved; hence Bordeaux does not prevent plant damage. These results may explain the reported effectiveness of Bordeaux under moist conditions in England and British Columbia, and its ineffectiveness under dry conditions in the United States, Europe, and Egypt. In contrast to Bordeaux, Parzate (2 lb./100 gal., plus above spreader) gave almost perfect control of rust, and, therefore, of secondary organisms, without host injury, in both localities.

Chemotherapeutic action of 8-quinolinol benzoate against Dutch elm disease. DIMOND, ALBERT E. When treated with 8-quinolinol benzoate, Dutch elm diseased trees suffer half as much as untreated trees. Beneficial effects are most pronounced when 0.1

per cent solutions are injected under pressure into the soil around feeding roots at the rate of 5 gal. per inch DBH. Surface watering over feeding roots is less effective and scattering of dry chemical followed by watering is ineffective. Treatment before leaf buds expand is without effect; in May and June treatment results in fewer foliage symptoms than in checks; and in August and September it reduces overwinter dieback. Trees treated in very early stages of infection are benefited most; those treated after involvement has become extensive are scarcely benefited. Although remedial action starts to disappear within 90 days and wears off completely in one year, differences between treated and untreated trees may be maintained by reapplying the chemical within six months to a year. Healthy trees may possibly be protected against natural infection when inoculum potential is low. Recovery of diseased trees is infrequent, but treated ones recover more often than untreated ones do.

Injury to cotton from 2,4-D weed-killer. DUNLAP, A. A. Fields of cotton in the Gulf Coast area of Texas, apparently injured by 2,4-D dust applied to neighboring rice fields in late May and early June, were kept under observation during the remainder of the growing season. The effects of the chemical on vegetative and fruiting parts of the plants were recorded in fields planted on different dates. No cotton plants were found that had died from the effects of the chemical in the affected fields. Young plants recovered from 2,4-D injury more rapidly than old plants. Rate and degree of recovery depended on such factors as age of plants, degree of injury, rainfall, and soil fertility. Owing to the high sensitivity of cotton to 2,4-D, minute traces of the chemical apparently can cause marked morphological changes. Evidence of impaired germination of the cotton seed has been obtained.

Effects of sugar concentration on the size of conidia produced by Helminthosporium victoriae. ELLIOTT, EDWARD S. As part of a more extensive study of factors influencing fructification of fungi, the effect of the carbohydrate content of culture media upon sporulation of *Helminthosporium victoriae* M. & M. has been investigated. Isolates from Victland oats (West Virginia) sporulated abundantly under most laboratory conditions. Conidia produced on medium containing 2 per cent sugar were approximately one-third the length and contained one-third as many cells as conidia produced on medium containing 0.1 per cent sugar. Within limits, the length of conidia and the number of septations are inversely proportional to the quantity of carbohydrate in the medium. Variation in kind and amount of nitrogen had no apparent effect upon the size of conidia. These results emphasize the need for caution in comparing spores produced in culture with those produced in nature. The diminutive conidia germinate as readily as normal sized conidia.

Investigations on the physiology and pathogenicity of Ustilago zeae. FELDMAN, A. W. A comparative physiological study was made on seven lines of *Ustilago zeae*. These lines included five haploids (17 D_4 and 10 A_4, lines of opposite sex, and three mutants of 17 D_4). The two solopathogens (410 n, a weak solopathogen, and 410 yy, a strong solopathogen) were obtained from a cross between 17 D_4 and 10 A_4. Differences among the lines were evident by: 1) pathogenicity; 2) proteolytic activity; 3) growth in liquid media; 4) effect of enzyme inhibitors; 5) effect of donators; and 6) respiration studies with seven carbohydrates, seven organic acids, and eight amino acids. Combination tests (employing manometric techniques) with lines of opposite sex as well as with the solopathogens indicated: 1) no stimulating substance(s) was formed when glucose was used as the metabolite; 2) stimulation was observed when two lines of opposite sex were combined with corn extract as the metabolite; 3) no stimulation resulted when two lines of the same sex were combined; and 4) stimulation substance(s) was obtained from culture extracts but not from galls or from the protoplasm of lines tested. It was possible to correlate the physiological activities of the lines and combinations with the pathogenicity ratings obtained by tests on corn inoculations over a two-year period.

Physiological effect of Ustilago zeae on corn. FELDMAN, A. W. The effect of *Ustilago zeae* on the host was studied by comparing the respiratory rates of the nodes from healthy and diseased plants, and of galls. The effect of a gall on the ear located at node 6 was a lowering of the respiration of all the nodes below the site of infection. No galls were produced by plants inoculated with line 410 n (a weak solopathogen) but nodal respiration was inhibited down to the fourth node. The aerobic respiration of the nodes from diseased plants was inhibited more than the anaerobic respiration of the corresponding nodes. It was possible to correlate the relative pathogenicity ratings for seven lines and combinations of *Ustilago zeae* to the Q_{O_2} values obtained at node 3 from plants inoculated with these lines and combinations. The R.Q. values for healthy nodes, nodes from diseased plants, and galls were 0.82–0.85, 0.68–0.73, and 1.20, respectively. Catechol and ascorbic acid increased the oxygen uptake of all three tissues. Potassium

bisulphite and sodium fluoride were without effect on any of these tissues, but malonic acid and sodium nitrite inhibited most the respiration of nodes from diseased plants. It was impossible to demonstrate any differences among the lines and combinations of *Ustilago zeae* in the physiology of the galls, or in the nodal response to inhibitors and donators.

Correlation between sugar beet crop losses and greenhouse determinations of soil infestations by Aphanomyces cochlioides. FINK, HARRY C. Soil samples were taken from 24 sugar-beet fields in the spring, prior to field planting. Treated (ethyl mercury chloride) seed was planted in these soil samples in the greenhouse. The percentage of seedlings infected in 30 days by *Aphanomyces cochlioides* was determined and recorded for each soil sample. The percentages ranged from 0 to 100. During August the loss in stand and the presence of diseased beets in each field was estimated as percentage loss of the crop. The estimates ranged from 0 to 95 per cent; three poor fields had been destroyed all or in part by growers. There was a correlation of 0.925 between percentage of seedlings killed by *A. cochlioides* in the greenhouse and field crop-loss estimates. Greenhouse determination of soil infestation by *A. cochlioides* offers promise as a means of predicting sugar-beet crop losses.

Hybridization between Ustilago hordei and U. bullata. FISCHER, GEORGE W. Several hybrids of *Ustilago hordei* × *U. bullata* were obtained, when one collection of *U. hordei* from *Elymus canadensis* and four races of *U. bullata* were crossed. These interspecific hybrids yielded F_1 spores which germinated in such bizarre fashion that F_1 spores had to be used to obtain the F_2 generation, rather than monosporidial cultures from F_1 spores. When *U. bullata* from *Bromus tectorum*, having verrucose spores with uniformly colored wall, was crossed with *U. hordei*, having smooth spores with wall lighter colored on one side, the F_1 and F_2 spores were echinulate and distinctly lighter colored on two sides. When *U. bullata* from *B. purgans*, having large (8–11 μ) verrucose spores with uniformly colored wall, was crossed with *U. hordei*, having smaller (5–8 μ), smooth spores with wall distinctly lighter-colored on one side, the F_1 spores were small (5–9 μ), minutely echinulate and with a tendency to be lighter colored on two sides. The F_2 generation segregated as follows: small (7–10 μ), echinulate, with a tendency to be lighter colored and more strongly echinulate on one side; small (6–9 μ), smooth, lighter colored on two sides; large (13–16 μ), punctate, with uniform wall; and large (11–12 μ), echinulate, with spores lighter colored and more strongly echinulate on one side.

Hybridization between Ustilago striiformis and U. bullata. FISCHER, GEORGE W. From pedigreed monosporidial cultures of opposite sex, 10 F_1 hybrids were obtained between *U. striiformis* forma *hordei* and two races of *U. bullata*, when *Agropyron trachycaulum* and *Elymus canadensis* served as common hosts. Without exception the F_1 sori were of the stripe type typical of *U. striiformis*, although in 9 of the hybrids sporulation was in the glumes and flag leaf rather than in the lower leaves. Germination of the F_1 spores was so bizarre that monosporidial cultures were not obtained and it was necessary to use F_1 spores as inoculum to obtain the F_2 generation. The percentage of infection obtained in the second generation was too low to permit the determination of genetic ratios of segregation. Three of the F_1 hybrids produced only stripe type sori in the F_2, one produced only head type, two segregated into both, and four failed to produce a second generation. The F_1 spores were minutely echinulate as compared with the prominent echinulations in *U. striiformis* and the rough verruculations in *U. bullata*. In the F_2 the head type sori contained verrucose spores, while the stripe type sori contained echinulate spores.

Host nutrition in relation to development of bacterial wilt of tomato. GALLEGLY, M. E. Bonny Best tomato plants were grown in constant drip sand cultures of 0.1, 0.5, 1, 2, and 3 times a basal salt solution and in cultures with the basal solution low and high in nitrogen, phosphorus, and potassium. Inoculations were made after 24 to 28 days' growth by dipping the washed roots in a concentrated cell suspension of *Bacterium solanacearum*. In concentrations of the balanced solution, disease development was greatest at the 0.1 level and decreased with an increase in salt concentration. Little difference was observed in the two highest solutions. In the unbalanced solution low in potassium, disease development was increased over that in the basal solution; while in the solution high in nitrogen, disease development usually was decreased. No correlation of disease development with growth of the host was observed.

Reaction of onion varieties to isolates of the pink-root organism. GORENZ, A. M., AND R. H. LARSON. Using a sand-culture seedling test, 48 lots of onion, including major standard varieties, were inoculated with isolates of *Phoma terrestris* from Louisiana,

Texas, California, Illinois, Iowa, Colorado, Utah, Massachusetts, and Wisconsin. Yellow Bermuda, Beltsville Bunching, and one lot of White Sweet Spanish were the most resistant. Crystal Grano, Ailsa Craig, and Autumn Queen were the most susceptible. In reaction to any given isolate, the varieties always assumed the same relation to one another in order of resistance and susceptibility, although some isolates were highly virulent and others mildly so. With the three most virulent isolates, one each from California, Colorado, and Louisiana, the Yellow Bermuda, Beltsville Bunching, and White Sweet Spanish seedlings were nearly all killed in the course of the test but nevertheless succumbed more slowly than the more susceptible varieties.

Investigation on the mechanism of fungicidal activity. GOTTLIEB, DAVID, AND DAVID DAVIS. Six materials were investigated to determine their effect on the respiration and growth of *Sclerotinia fructicola.* The fungicides were used at concentrations of 0.1, 1.0, 10, 100, and 1000 p.p.m. in Blakeslee's 230 medium. Growth was determined after a 48-hour period and respiration was measured in a Warburg manometer for three hours after the addition of the compound. The effects of all compounds were not the same, since growth was more readily inhibited than was respiration with some of the materials but not with others. Thus 8-quinolinol sulfate and copper-8-quinolinolate inhibited growth almost completely at concentrations as low as 0.1 p.p.m. whereas no inhibition of respiration could be measured with 10 p.p.m. No inhibitory effects were observed with 5,7-dinitro-8-quinolinol at any of the concentrations used. Isothan Q15 both prevented growth and inhibited respiration almost completely at 10 p.p.m. Copper sulfate $5H_20$ exerted no deleterious effect on either growth or respiration in Blakeslee's medium until 1000 p.p.m., but when the respiration of washed mycelium in a 4 per cent glucose solution was measured, inhibition of respiration was observed at 1.0 p.p.m. Variable results were obtained with 2,4-dinitrophenol.

Preliminary observations and experimentation on relation of insects to purple-top of potatoes in Minnesota. GRANOVSKY, A. A., AND A. G. PETERSON. Purple-top wilt of potato, occasionally serious in Minnesota, was of minor importance in 1947. The symptoms of purple-top wilt, as occurred in Minnesota are differentiated from similar symptoms associated with other diseases. Although the symptoms agree with those reported by Leach for purple-top wilt, the definite cause of the disease has not been verified. During 1947, purpling of foliage in most fields was the result of rhizoctonia stem cankers and blackleg infections rather than purple-top wilt. Usually purpling symptoms caused by these other diseases are attributed to purple-top wilt. Efforts to produce typical purple-top wilt on potatoes in outdoor cages and greenhouse have thus far been unsuccessful. Insects experimentally used: *Macrosteles divisus* from aster-yellows-infected asters; from barley; and from wild host plants; also such insects as *Polyamia inimica, Adelphocorus lineolatus, A. rapidus, Lygus oblineatus, Miris dolabratus, Stenotus binotatus,* and *Capsus atratus.* Periodic insect surveys of potato fields were made in various sections of the State during the season. The only appreciable differences in insect and disease occurrence in so-called purple-top areas have been greater abundance of *Lygus* and *Adelphocorus* spp. and increased incidence of aster-yellows on carrots without increased populations of *M. divisus.*

A pod-distorting strain of the yellow-bean mosaic virus. GROGAN, RAY G. In 1946 bean plants of Refugee varieties highly resistant to *bean virus 1* were found in Wisconsin fields, which showed leaf symptoms of yellow bean mosaic (*bean virus 2*), but were more severely stunted than typical, and bore severely distorted pods. The common strain of *bean virus 2* infected all varieties tested, causing stunting and chlorotic mottling of the leaves. The virus from pod-distorted plants did not infect Great Northern UI 59, UI 123, and UI 81. It caused top necrosis on Great Northern UI 1 and UI 15, Michelite, Stringless Blue Lake, Scotia, Tendergreen, Bountiful, Potomac, and McCaslan, and its host range was much narrower than that of the common strain. Its physical properties were: heat inactivation, 58–60° C.; longevity *in vitro,* 32 hours; dilution end point, 1–2000. Richards and Burkholder's strain of *bean virus 1* and the common strain of *bean virus 2* both protected against top necrosis on Michelite, while southern bean mosaic (*bean virus 4*) and Whipple and Walker's strain 14 of *cucumber virus 1* gave no protection. The pod-distorting virus is considered a strain of *bean virus 2* and as being related to Richards and Burkholder's strain of *bean virus 1.*

The relation of common bean mosaic to black root. GROGAN, RAY G. Certain varieties of string bean derive their high resistance to common mosaic and greasy pod from Corbett Refugee. Ordinarily, plants of these varieties show no symptoms when inoculated with *bean virus 1,* and the virus is not recovered from such plants. It was noted that rare plants, inoculated by rubbing with the virus of common mosaic or that of greasy pod, developed extreme necrosis symptoms. When plants were inoculated by

approach graft with susceptible Stringless Green Refugee plants (which were inoculated after the graft union took place) a high percentage of plants developed necrosis similar to that described for black root. When plants were inoculated by hypodermic injection similar results were secured. If plants were so inoculated in the early-pod stage, necrosis of pods characteristic of black root followed. It is concluded that black root is an expression of common bean mosaic by plants that are highly resistant to the virus. Varieties of bean, deriving their resistance from Robust or Great Northern varieties, and in which resistance is distinct genetically from that of Corbett Refugee, showed no necrosis or black root when inoculated. All bush-bean varieties tested that were tolerant or susceptible to *bean virus 1* remained free from black root.

Apple rust controlled by airborne application of Fermate. GROVES, A. B. Seven-acre plots of thirty-odd-year-old York and Winesap apple trees received airborne applications of spray concentrates beginning with the pink spray and continuing through the first cover spray when applications were discontinued after a freeze-out. Fungicides used in the concentrate formulae included liquid lime-sulfur, Puratized Agricultural Spray, and Fermate, plus various adjuvants. Application was at the rate of 8 gallons per acre; 7½ oz. of Fermate was used per gallon. A single Fermate application made on May 11 reduced foliage infection from an average of 17.94 lesions per leaf on the poorest plot to 1.05 per leaf, a reduction of over 94 per cent. The application followed a favorable rust infection period of six days duration by four days, and was followed by another period of two days duration after two days.

Compatibility of organic fungicides with acaricides and DDT. GROVES, A. B. Combinations of organic fungicides plus DDT with the acaricides DN–111, Compound 714, Genicide, and hexaethyltetraphosphate were applied to Delicious, Grimes, and York apples to determine their compatibility in terms of fungicidal effectiveness and safety. Fermate and Bioquin gave fair scab control and were unaffected by all acaricides except Genicide, which added to their effectiveness. Phygon and Compound 341 gave good scab control and were unaffected by combinations. Scab control with Puratized was poor and variable, its effectiveness being reduced by hexaethyltetraphosphate and fortified by Genicide. Fermate and Phygon gave excellent rust control in all combinations. Puratized gave fair rust control, Genicide increased its effectiveness while both hexaethyltetraphosphate and Compound 714 destroyed its value against rust. Bioquin was ineffective against rust and although Compound 714 increased its effectiveness some, best results with Bioquin were obtained when used with DN–111 or Genicide. Compound 341 gave fair rust control and was unaffected by all combinations. All organic fungicides tested proved non-injurious when used with DDT alone. All combinations with Genicide were unsafe, that with Compound 341 causing the least injury. Phygon plus DN–111 caused fruit and foliage injury. Fruit set was too variable to secure complete data.

Three viruses of canning pea. HAGEDORN, D. J. Three apparently undescribed viruses of canning pea, *Pisum sativum*, were isolated from plants in Wisconsin pea fields. One virus produced mottle of the leaves; another caused streaking of the stem; the third brought about stunting of the plant. Host range tests indicated that all three viruses are limited to the Leguminosae. The mosaic and stunt viruses were transmitted by pea aphid (*Illinoia pisi*), but attempts to transfer the streak virus by this means yielded negative results. The mosaic virus was infective to *Melilotus alba, M. officinalis, Trifolium pratense, T. incarnatum, Phaseolus vulgaris, Vicia faba, Lathyrus hirsutus, Trigonella foenum-graecum*, and *Pisum sativum* var. *arvense*. It was not transmitted through seeds collected from diseased peas or beans. It tolerated dilution of 1:10,000 and was inactivated *in vitro* within 24 hours. The streak virus was infective to *M. alba, Trifolium pratense, T. incarnatum, T. hybridum*, and *L. hirsutus*. It tolerated dilution of 1:1,000,000 and remained active *in vitro* for seven days. Hosts of the stunt virus included *T. pratense, T. incarnatum, Trigonella foenum-graecum*. It was slightly infectious at 1:10,000 dilution and was inactivated *in vitro* at about 48 hours.

Cure of crown gall with antibiotics. HAMPTON, JACK E. Through the use of penicillin and streptomycin, both commercial and locally produced crude, numerous cures of crown gall have resulted. Usually the galls were treated by applying to them a cotton-wool pad saturated with the antibiotic. Streptomycin is somewhat more efficient than penicillin, but more than one treatment with either has seldom been necessary. The antibiotics have also been used in immersions of galled roots and by hypodermic injections. Treated galls have been cured on *Bryophyllum pinnatum, Lycopersicon esculentum, Ricinus communis, Prunus domestica* (Green Gage Plum), *P. salicina* (Satsuma Plum), *Pyrus communis* (Bartlett Pear), *Prunus cerasus* (Duke Cherry), *Ligustrum lucidum*, and other plants. Galls treated on these plants were either aerial or sub-

terranean, soft or hard, on the stems, taproots, and secondary roots. Use of antibiotics on crown gall should prove particularly valuable in nurseries.

Victoria blight of oats in Kansas. HANSING, E. D., HURLEY FELLOWS, C. O. JOHNSTON, AND A. L. CLAPP. In 1946 Victoria blight caused a loss of 1 per cent. In 1947 the disease was generally distributed on susceptible varieties and caused a loss of 30 per cent in northeastern Kansas, 20 per cent in southeastern Kansas, 5 per cent in central Kansas, and a trace in western Kansas. Osage, Neosho, Boone, and Tama were susceptible, while Kanota and Fulton were resistant. Cherokee, Nemaha, and Clinton were recommended for 1948. These are resistant to Victoria blight, smut, and the crown and stem rust races found commonly in Kansas. Victoria blight was favored by high soil temperatures (optimum 22° to 26° C.) and by moderately high moisture. No symptoms of the disease were observed on Neosho oats grown in soil at 10° and 13° C., but when these plants were transferred to 23° C. the disease developed rapidly. Seed treatment was effective in controlling the disease in sterile soil, but only partially effective in infested soil. Crop rotation was partially effective in controlling Victoria blight. In eastern Kansas, when susceptible oat varieties were preceded by the following crops, percentages of loss occurred as follows: susceptible oat varieties, 38; corn, 27; clover and meadow, 26; soybeans, 24; wheat, 18; and resistant oats, 4.

Field inoculation technique with Aphanomyces root rot of garden pea. HARE, WOODROW W., AND DARRELL G. WELLS. Natural infection by the root rot organism, *Aphanomyces euteiches* Drechsler, is extremely variable from year to year in Wisconsin. To test possibilities of artificial inoculation of field plots two varieties of garden pea (*Pisum sativum* L.) were planted in 1947 according to a split-plot design. The Pride variety, because of observations indicating possible tolerance to the disease, was compared to highly susceptible Wisconsin Perfection. Three applications of inoculum of *A. euteiches* were placed on separate rows at different times with one row left as check. Plants were classified according to disease reaction 76 days after planting, and disease indices were calculated. There were no significant differences between varieties or interaction of varieties and treatment. Differences between treatments were highly significant. Inoculations made before planting and when the peas were 6 to 8 inches high produced significantly more disease than inoculation when the peas were in late blossom. Late blossom inoculation was significant over the check.

Stem rust of wheat in Mexico. HARRAR, J. G., AND N. E. BORLAUG. As previously reported, the tendency for the establishment of all of the important physiologic races of *Puccinia graminis tritici* in all wheat-growing areas of Mexico is increasingly evident. Race 38 is regularly found in both northern and central Mexico although not reported from Sonora in the northwestern part of the country. Race 17 is found in all three regions as is race 56. Races 59, 59–A, and 19 are still restricted to central Mexico. It is probably not feasible to consider that varieties may be selected for the several wheat-growing areas of Mexico on the basis of their resistance to predominant races found in those regions, but rather is it necessary to consider that improved wheats for Mexico must be developed with reference to all the important races found in the country. At the present time there are available for increase, selections from varieties of Kenya, Trigo Supreme × 41–116, and Frontaira × 41–116, and selections from crosses of Newthatch × Marroqui, Kenya × Aguilera, and Mida × Pelón Colorado, which in test give promise of a high degree of resistance to the predominant physiologic races found in Mexico.

Resistance in onion to smudge, neck rot, and black mold. HATFIELD, W. C., AND J. H. OWEN. Twenty-two varieties of onions were tested in an attempt to correlate disease incidence with certain characteristics of the varieties. Using a bioassay, little correlation was found between the fungicidal and fungistatic action of the expressed juice and its vapor and disease incidence of smudge (*Colletotrichum circinans*). A definite correlation existed between certain scale characters and smudge incidence in colored varieties. Those varieties with poor outer scale adherence were more susceptible than those with tight, well developed scales. This does not apply to the white varieties where absence of color leads to susceptibility. The incidence of neck rot (*Botrytis allii*) was definitely correlated with pungency, those varieties highly pungent having lower disease incidence than the milder varieties. Incidence of black mold (*Aspergillus niger*) was correlated with bulb color differing from smudge in that colored varieties were more susceptible than the white ones. The parasitic action of the three organisms varied with their tolerance to the antibiotic substances present in expressed juice of the succulent scales. *B. allii* was the most aggressive and the least affected by the antibiotic substances. *C. circinans* was less aggressive and more affected. *A. niger* was least aggressive and most affected by the antibiotic substances.

Pathogenicity of single spore isolates of the fusarium-wilt organisms of muskmelon and watermelon. HENDRIX, J. W., E. P. DU CHARME, AND HARRY MURAKISHI. Previous workers have attributed watermelon wilt and muskmelon wilt to forms 1 and 2 of *Fusarium bulbigenum* var. *niveum*, respectively, and have found no exceptions to strict host-pathogen specificity. Further research by the writers has shown that some isolates from wilted muskmelon plants cause severe pre-emergence killing and wilting of watermelon seedlings in the greenhouse. Some isolates from watermelon parasitize muskmelon. For example, certain monosporous isolates from muskmelon caused as much as 40 per cent reduction in stand of watermelon while similar losses resulted when muskmelon seedlings were grown in soil infested with isolates from wilted watermelon plants. On the original host plants, different isolates caused from 0 to 100 per cent killing. In general, the virulence of an isolate was greater on its original host than on the second host plant; nevertheless, several isolates from watermelon caused a more severe reduction in stands of muskmelon than of watermelon.

Wilt epidemiology and resistance in the mimosa tree. HEPTING, GEORGE H., AND E. RICHARD TOOLE. Within the 12 years since the fusarium wilt of *Albizzia* was discovered, it has spread widely until now it is known in 76 counties from Maryland to Alabama. At Morganton, North Carolina, where the wilt appeared on one city block in 1943, trees were dead or dying on 232 blocks by 1947. Search for wilt-resistant mimosas was begun in 1939. Six hundred and thirty-two seedlings were grown from seed collected from Maryland to Louisiana. After several inoculations, the remaining seedlings were planted in infested soil. Twenty of them have remained wilt-free for 5 years, while neighboring volunteers have died. Eight stem cuttings rooted from these trees and 17 from some random volunteers were inoculated at one time. All volunteer cuttings died of wilt the first year, but none of the cuttings from the resistant selections died. Rooted cuttings from trees selected at random from other locations have also proved to be susceptible. Thus, certain individual mimosa trees appear to be wilt resistant.

The dithiocarbamate fungicides and the control of downy mildew disease of cucumbers and cantaloupes. HEUBERGER, J. W., AND L. P. NICHOLS. Research during 1946 and 1947 on the use of Zerlate (zinc dimethyl dithiocarbamate), Fermate (ferric dimethyl dithiocarbamate), Dithane Z–78 and Parzate (zinc ethylene bisdithiocarbamate), Dithane D–14 + zinc sulphate–lime (zinc ethylene bisdithiocarbamate), and manganese ethylene bisdithiocarbamate as sprays at 1½–100 active ingredient concentration for the control of downy mildew on cucumbers (pickles) and cantaloupes, has shown the following. *Cucumbers:* Zerlate, Parzate, and Dithane Z–78 were as effective in control as Bordeaux and the fixed coppers, increased early and total yields more than the copper compounds, and were noninjurious when properly formulated; Dithane D–14 + zinc sulphate–lime was not so effective as Zerlate, Parzate, and Dithane Z–78; Fermate was injurious under certain conditions (hot, dry weather); manganese ethylene bisdithiocarbamate was injurious. Yield increases from Zerlate, Parzate, and Dithane Z–78 were respectively, 48, 29, and 50 per cent in 1946 and 54, 81, and 71 per cent in 1947. *Cantaloupes:* Zerlate, Dithane Z–78, Parzate, and Dithane D–14 + zinc sulphate–lime were equal in control to Bordeaux and the fixed coppers and gave higher yields. Fermate and manganese ethylene bisdithiocarbamate were injurious. When similar materials were compared as sprays and dusts in 1947, the dusts give higher yields under the hot, dry weather conditions prevailing in late July and August.

Induced wilting of tomato cuttings by synthetic polymers of varying molecular weight. HODGSON, ROLAND, W. H. PETERSON, AND A. J. RIKER. Studies with certain toxic water-soluble polysaccharides have suggested a direct relationship between the molecular weight (range 1,000 to 5,000) and the wilt-inducing potency on tomato cuttings. Such a relationship was sought with other water-soluble polymers available in several molecular-weight ranges. Solid polyethylene glycols (0.2 to 0.4 per cent aqueous solutions) induced severe wilting and necrosis of leaflets; the symptoms varied somewhat with the different compounds but were similar to those caused by a glucosan from crown-gall bacteria and certain other polysaccharides. A direct relationship between molecular weight (range approximately 1,500 to 9,000) and wilt-inducing activity was found, the largest being most toxic. This suggests that the wilting mechanism may be mostly physical. Evidence for an accumulation of the toxic agent in or near the injured areas was sought by analyses of cuttings treated with polyethylene glycols. Of the glycol taken up, 87 to 97 per cent was recovered; of this, 81 to 91 per cent was found in the leaflets. Wilted portions of leaflets contained more glycol than non-wilted portions, showing a greater accumulation in the region of the injury.

A new type of resistance to spotted wilt. HOLMES, FRANCIS O. Spotted-wilt virus isolated from a recent outbreak of disease in dahlia and tomato in New Jersey was found

to be capable of overcoming the characteristic resistance of the Pearl Harbor tomato, which is adequate for protection from spotted wilt in the Hawaiian Islands. A new and locally applicable type of resistance was found in a tomato derived from Argentina. This new type of resistance proved effective both in greenhouse and field tests. It showed monofactorial inheritance in hybrids with the susceptible variety Rutgers. The new type of resistance together with the only formerly known type, that in the Pearl Harbor tomato, may be expected to serve a dual purpose, of facilitating control of the disease and of indicating the geographical distribution of differing strains of spotted-wilt virus.

Evidence of parasitic activity of Actinomyces scabies on seedling roots. HOOKER, W. J., AND J. E. SASS. It was previously reported that seedling roots of a number of plants developed severe necrosis when grown in soil-water agar artificially infested with *Actinomyces scabies* whereas roots of the same species showed no necrosis with cultures nonparasitic to potato. Necrosis of a comparable type, although less severe, developed on soybean and wheat roots grown in quartz sand artificially infested with *A. scabies*. There was no evidence that such necrosis was produced by a water soluble substance secreted by *A. scabies* on potato-dextrose agar. Sectioning demonstrated *A. scabies* filaments abundant in the rhizosphere and in the epidermal and cortical cells of these roots grown in agar even though the root was not noticeably necrotic. *Actinomyces* spp. were demonstrated by dilution plate counts to be appreciably more abundant in the rhizosphere of field-grown oat roots than in soil in which there were no roots. Roots of Clinton oat plants grown in the peat soil in northern Iowa were sectioned and the filaments of an actinomycete were demonstrated in the tissue.

Some histological features of potato stem necrosis associated with Actinomyces scabies. HOOKER, W. J., AND J. E. SASS. Of the varieties of potato tested to date, those known to be susceptible to tuber scab (*Actinomyces scabies*) exhibited considerable necrosis of stems resulting from infection by the scab organism, whereas varieties resistant to tuber scab were likewise resistant to stem necrosis. When plants were grown in the greenhouse in sterilized peat soil artificially infested with *A. scabies*, infection was often established in the stems through unwounded surfaces away from stolons and secondary roots. In this type of lesion, the susceptible varieties, Red Warba, Cobbler, Pawnee, Katahdin, and Chippewa, showed either no periderm or at most a poorly developed, discontinuous periderm, with actinomycetous filaments abundant in the cortical cells. The stems of the resistant varieties, Menominee, Cayuga, and two unnamed clones, showed well-developed and continuous periderm with sparse intracellular mycelium. This relationship did not hold consistently with resistant and susceptible varieties naturally infected in the field. Where stem infection in the field took place through natural wounds such as longitudinal splits or at points of emergence of stolons and secondary roots, periderm formation was generally poor, even in resistant varieties, and mycelium was often abundant.

A spiral design for the field assay of pesticides. HORSFALL, JAMES G., SAUL RICH, AND NEELY TURNER. Single-row plots are arranged in a continuous spiral row. Planting is simplified by attaching the planter or marker to a cable that winds around a fixed vertical drum anchored at the center. The planter progresses inwardly at a uniform rate allowing three-foot intervals between laterally adjacent plots. A roadway is left for placing the trailer-mounted power sprayer at the spiral center, near the water supply and drainage pipes. An overhead, rotatable, horizontal aluminum pipe carries the spray material to a length of rubber hose attached to a hand-carried, three-nozzle spray boom. The sprayer has two tanks for simultaneous spraying and mixing. Ten-foot plots of nonstraggling plants are randomized in each quadrant, making four replicated wedge-shaped blocks. The hand-carried spray boom is flexible, the power pump untiring; the circular route of travel saves a return empty trip; and the water supply and drainage arrangements save work and time in loading. Drift error is negligible. Spring-loaded check valves, ahead of the spray nozzles, prevent drizzle between plots. Inoculation is aided by a rotating overhead irrigator, controlled at night by a timing apparatus.

Root-rot complexes of tobacco and small grains in Virginia. JENKINS, WILBERT A. Studies of several root rots of tobacco and cereals in Virginia offer proof that feeder root mutilation is a primary factor in initiation of the diseases. Feeder root mutilation is accomplished through the feeding habits of various members of the meadow nematode group. Representatives of two nematode genera, *Pratylenchus* and *Ditylenchus*, cause primary pathologic symptoms in addition to serious mutilation while several representatives of other genera appear principally to open up the tissues to invasion by fungus and bacterial soil associates. These studies have reference to brown and black root rot of tobacco as well as certain unclassified troubles symptomatically characterized by stunting,

malnutrition, and low production. The studies on small grains included "winter" injury of wheat and barley and certain unclassified diseases of wheat, barley, and oats whose principal symptoms were stunting, foliage bronzing, and unproductiveness. Such factors as relatively low soil temperatures and relatively high calcium levels distinctly favored nematode population increase and severity of feeder root mutilation of all groups. Higher soil temperatures and moisture in the later season definitely favored root decays, depending on the specificity of the bacterial and fungus associates in the rhizosphere.

Strains of flue-cured tobacco resistant to black shank (Phytophthora parasitica var. nicotianae) and tolerant to certain root-rot complexes. JENKINS, WILBERT A., Crosses were made in 1941 between two resistant tobaccos and a strain of Yellow Special. The resistant parent lines were developed in North Carolina from a cross between Florida 301 × Warne, with multiple backcrosses to Warne (later released as Oxford 4) and a cross between Florida 301 × White Stem Orinoco, with multiple backcrosses to W.S.O. (later released as Oxford 3). The resistant parents were about 80 per cent resistant to black shank, but low in quality and yield and susceptible to root-rot complexes prevalent in Virginia. The Yellow Special parent was susceptible to black shank, but of superior quality and yield and highly tolerant to several common root-rot complexes. Following three seasons of mass selection and three seasons of pure-line selections from progenies of these crosses, selected lines have been fixed for type. These are 95–98 per cent resistant to black shank under field conditions, of excellent quality and yield, and highly tolerant to several of the root-rot complexes found in Virginia. Throughout the period of selection, test-plot soils were heavily infested with black shank and each individual plant was inoculated with black shank material each season. Available at present are strains representing 4 leaf types. All strains were evaluated in Virginia and North Carolina last season.

Increase of virus infectivity by certain chemicals. JOHNSON, JAMES, AND PHOEBE A. GREEN. In 1935 Stanley listed 17 chemicals that increased the infectivity of the ordinary tobacco-mosaic virus. We have tested these chemicals by somewhat modified methods. The sensitive *tabacum-glutinosa* hybrid was used as a local-lesion test plant. Inoculations with the virus-chemical mixtures were made at once and after different intervals of storage. The infectivity with five chemical treatments, involving 87,324 local lesion counts with controls based at 100, were: sodium nitrate, 190; calcium chloride, 166; dipotassium phosphate, 138; sodium thiosulphate, 121; diethyl acetate, 118. The highest increase (226) was obtained with sodium nitrate after immediate inoculation. Increases as high as some reported by Stanley were not obtained. The other chemicals listed by Stanley either did not yield increases or they were too small in amount to be significant. The increased infectivity declined very slowly in storage and was retained for as long as 30 months at dilutions of 1–1000, suggesting some "preservative" value of the chemicals mentioned. The evidence secured did not indicate that increased infectivity is due to a change in hydrogen-ion concentration. It seems more likely that these chemicals favor a higher dispersion of the virus particles in the inoculum.

A yellow-mosaic virus associated with Eryngium mosaic. JOHNSON, JAMES, AND EDITH M. HEIN. Further studies have been made on a yellow-mosaic virus from a complex in *Eryngium aquaticum* L. This component produces a uniform chlorosis over older leaves of Havana tobacco plants, with occasional broad chlorotic spots or patterns on younger leaves. It is difficult to separate this virus or strain from the Eryngium-mosaic virus, although the latter may be readily freed of the yellow type. Property determinations indicate that the yellow-mosaic virus is somewhat less tolerant than is the Eryngium-mosaic virus, whose properties are similar to those of *Cucumber virus 1.* Furthermore, the yellow-mosaic virus was not transmitted by the peach aphid and did not yield symptoms on tomato, which is susceptible to the Eryngium-mosaic virus. The best differential host for the yellow Eryngium-mosaic virus appears to be White Burley (var. Judy's Pride) tobacco. On this host, inoculative and systemic chlorotic spots and necrotic lesions occur which, by repeated transfer, are helpful in freeing the virus from the Eryngium-mosaic virus. The chief interest in yellow-mosaic viruses centers around their origin and relationship to other viruses with which they are normally associated. Their ability to destroy chlorophyll or to interfere with its normal production is characteristic and significant.

A new virus disease of Ladino clover. KREITLOW, K. W., AND W. C. PRICE. A virus disease of Ladino clover (*Trifolium repens* var. *giganteum*) characterized by yellow mottling of leaves and retardation of growth, occurs extensively in the Northeastern United States. The disease is caused by a virus which is readily transmitted mechanically

to healthy Ladino clover. The virus was also transmitted to other species of *Trifolium*, to several species of *Melilotus*, and to *Pisum sativum*, *Phaseolus vulgaris*, *P. lunatus*, *Soja max*, *Zinnia elegans*, *Antirrhinum majus*, *Capsicum frutescens*, *Petunia hybrida*, *Nicotiana tabacum*, *N. glutinosa*, *N. rustica*, and *Vinca rosea*. Virus was recovered from each of these species. Typical chlorotic areas were produced in the leaves of many of the hosts tested. Symptoms in other hosts consisted of extensive mottling, downward curling of leaves, and severe stunting. Reddish-brown local lesions were produced within 48 hours when juice from infected plants was rubbed on leaves of *Phaseolus vulgaris*. The host range and properties of the virus resemble those of alfalfa mosaic virus but the symptoms are different from any known strain of the latter.

Autogenous necrosis in interspecific tomato hybrids and its relation to the breeding of tomatoes for resistance to Cladosporium fulvum. LANGFORD, ARTHUR N. Following crosses between the red currant tomato, *Lycopersicon pimpinellifolium*, and varieties of the cultivated tomato, *L. esculentum*, immune *esculentum*-like hybrids normally develop a severe autogenous necrosis, manifest chiefly in leaf-spotting, unless, along with Cf_{PI}, the dominant *pimpinellifolium* factor *Ne* is also incorporated in the hybrid. Susceptible plants show no necrosis, regardless of their genotype at the *ne* locus. Necrosis is due to the interaction among Cf_{PI} (located on chromosome 4), *ne* (located on chromosome 1) and, probably, other *esculentum* genes. Evidence has been obtained showing that one or more genes on chromosome 7 may, in progenies resulting from selfing, prevent the development of necrosis in immune plants of genotype *ne/ne*. Striking seasonal variations in the severity of necrosis have been observed: the artificial lowering of the incident light intensity greatly reduces the severity of necrosis during the summer. Although the discovery of races of *Cladosporium fulvum*, to which such varieties as Vetomold (Cf_{PI}/Cf_{PI}; *Ne/Ne*) are completely susceptible, has complicated breeding programs and initiated searches for additional resistance factors, the widespread distribution of races 1 and 2 suggests that the incorporation of the factor Cf_{PI} in resistant varieties will still be desirable. The dominance of the immunity factor and of the factor conditioning freedom from necrosis make the backcross method of developing new, necrosis-free varieties a convenient one.

Greenhouse tests of an antibiotic substance as a protectant spray. LEBEN, CURT, AND G. W. KEITT. In greenhouse tests ethanol solutions of an unidentified antibiotic substance produced by a species of *Streptomyces* were effective as protectant sprays against apple scab. Under certain conditions these sprays caused marked phytotoxicity due to the ethanol. In the present work there was no injury of apple, tomato, or pea plants when the antibiotic material was carried in 25 per cent ethanol in water or in water alone. With the more effective of these preparations, containing 3–6 arbitrary units of the antibiotic substance per ml. as determined by plate assay with *Glomerella cingulata*, complete control of apple scab and early blight of tomato was obtained in greenhouse experiments. Artificial rain tests indicated that the active material resisted washing on apple or tomato foliage. When apple leaves were inoculated with *Venturia inaequalis* four days after application of the antibiotic spray there was some lessening in the effectiveness of control. Yields of 3–5 units of the active material per ml. of the culture filtrate have been obtained from tank fermentations.

Comparative cultural studies with Thielavia basicola and Thielaviopsis basicola. LUCAS, G. B. The ascomycetous fungus *Thielavia basicola* has been reported as the perfect stage of the tobacco black root-rot fungus *Thielaviopsis basicola*. Perithecia and ascospores of *Thielavia basicola* have been seen on diseased tobacco roots in association with chlamydospores of *Thielaviopsis basicola*. To date it has not been possible to show that either of these forms would produce the other. In the absence of such data isolates of both were obtained and grown on various media and under different conditions to see how each would react. On Richards agar minus sucrose, with filter paper as a carbon source, *Thielavia* grew rapidly, *Thielaviopsis* grew little or not at all. On carrot slices *Thielavia* grew slowly or not at all, *Thielaviopsis* grew rapidly. *Thielavia* grew on potato or carrot agar at pH 3, *Thielaviopsis* did not. *Thielavia* grew at 37° C., *Thielaviopsis* did not. Pathogenicity tests on tobacco with *Thielavia* were negative, whereas *Thielaviopsis* was consistently pathogenic. When the two forms were grown together on various media, no mating reaction was evident. These data are offered as further evidence that *Thielavia basicola* is not the perfect stage of *Thielaviopsis basicola*.

Results from co-operative trials with seed disinfectants on flax. MACHACEK, J. E. As a part of the program of investigations sponsored by the American Phytopathological Society's Seed Treatment Committee, experiments involving a comparison of the newer seed disinfectants with respect to their effect on germination and yield of flax

were carried out at 14 stations in Canada and the United States. The results showed significant increases in germination and yield at some stations but not at others. Seed disinfection was more beneficial to mechanically injured seed than to uninjured seed. Of the disinfectants tried, New Improved Ceresan, applied at the rate of 1 to 2 oz. per bushel, generally gave the best result.

Argentine rape as an artificial weed in experiments with seed disinfectants. MACHACEK, J. E., AND H. A. H. WALLACE. The oil crop Argentine rape (*Brassica napus* L.) was used to infest experimental plots with an artificial weed with the object of simulating the weedy conditions of the ordinary farm. In plots thus infested the yield of flax or wheat was found to be almost proportional to the rate of seeding. In weed-free plots, where yields were in general much higher, the increase in yield was not proportional to the increased rate of seeding—the yield at first rising rapidly to a peak and then falling off with further increase of seeding rate or, in some cases, remaining at the peak level. The implication of this finding is that seed disinfection, by increasing density of stand, may be more profitable under weedy conditions than under weed-free conditions, and that a count of seedlings may be more reliable than yield as an index of seed disinfectant values.

Changes induced in Sclerotinia fructicola on copper sulphate media. MADER, E. O., and C. L. SCHNEIDER. The tolerance of 5 monoascospore isolates of *Sclerotinia fructicola* to copper sulphate in potato-dextrose agar was determined. All isolates tolerated copper sulphate concentrations up to 7,000 p.p.m. Sporulation was increased at concentrations below 1,000 p.p.m. but suppressed at concentrations of 5,000 p.p.m. or higher. After having grown for 18 weeks on 5,000 p.p.m. all isolates tolerated 10,000 p.p.m. Isolates that had grown for 18 weeks on media containing 5,000 p.p.m. of copper sulphate were transferred to non-copper media and tested for copper tolerance and cultural characters at the end of 14 weeks and 42 weeks. Some cultures reverted to their parental type in appearance and had lost their acquired tolerance for copper. Other isolates, however, did not revert to the parental cultural type and maintained a high tolerance for copper sulphate. These variants differed in ability to rot fruit.

New fungicides for control of narcissus basal rot. MCCLELLAN, W. D. New Improved Ceresan and 2 per cent Ceresan are both effective in controlling narcissus basal rot caused by *Fusarium oxysporum* f. *narcissi* but frequent flower injury results from their use. Hence an effective but safer treatment is desired. Five-minute after-harvest plus pre-planting dips of King Alfred narcissus bulbs either in Mersolite 8 (phenyl mercuric acetate) at the rate of one pound in 800 gallons, or in Puratized Agricultural Spray (phenyl mercuri triethanol ammonium lactate), one part of the active ingredient in 10,000, in 1945 and 1946, have given control of basal rot equal to the standard New Improved Ceresan treatment but without the flower injury. Rot has not been controlled so well when the bulbs were dusted with Arasan or with Spergon but bulb yields have been about the same as yields following treatments with mercurials. Beginning with 200 bulbs (weighing 284 oz.) per treatment in 1945 the bulb yields in 1947 after two years of treatment were: New Improved Ceresan, 531.5 oz.; Arasan, 523; Spergon, 490; Tersan, 393.5; Mersolite 8, 522.5; Puratized Agricultural Spray, 435.5; and untreated, 109.5 ounces. Similar results were obtained from treatments begun in 1946. A number of other materials have given either poor control or severe flower injury.

Etiology and control of begonia bacteriosis. MCDONOUGH, E. S. The leaf spot disease of begonia caused by the bacterium that has been referred to as *Xanthomonas begoniae* has not been satisfactorily controlled by modification of cultural practices when the plants were grown in large numbers under economically feasible greenhouse conditions. Studies on the etiology have made possible an efficient eradication procedure. Bacteria obtained from drops of sterile water which had been placed on diseased leaf regions were atomized on leaves of healthy Melior begonia plants, kept under conditions of high humidity. The plants were then sprayed with water. Only leaves atomized from the top showed symptoms typical of the naturally infected leaves. From this and other evidence it was assmed that infection generally took place by splashing during watering or by movement of the bacteria from leaf to leaf in contact with the rooting medium. Bordeaux mixture has controlled the disease, whereas other bactericides used have proved injurious to the host plant or have shown no superiority. Dipping the cuttings in Bordeaux and keeping the tops of the leaves covered during a season has resulted in practical elimination of the disease.

Relation between the condition of the seedling root system of corn and the grain. MEREDITH, CLIFFORD H. The seedling root system of the corn plant never attains a very

great length, so that it can usually be recovered for observation purposes. The mature seedling roots become rather woody and can be recovered even after over-wintering in the soil. A study was made of the condition of the seedling root system and the weight of grain produced by individual corn plants. One hundred plants taken at random from the field were measured to find, (1) circumference at the base of the stalk, (2) length of the primary root, (3) number of secondary roots, (4) length of the longest secondary root, and (5) weight of the grain produced. It was found that there was a relation between the yield and the length of primary and secondary roots. The cause of the poor condition of some of the seedling roots was apparently fungus attack at an early stage of growth. It appears that a plant attacked by a root pathogen at an early stage does not recover sufficiently to produce a normal yield. The results of these studies indicated that the condition of the seedling root is closely related to production of grain.

Root nodulation of Holland Jumbo strain peanut grown from seed treated with a fungicide. MILLER, LAWRENCE I. In a greenhouse test Spergon (tetrachloro parabenzoquinone) applied at the rate of 3 ounces per 100 pounds of hand-shelled seeds had no effect on root nodulation as measured by number, type, and location of nodules. This was true both when the Speregon-treated seed was inoculated with 2 commercial preparations of *Rhizobium leguminosarum* (mixed cultures in a humus base) and when left uninoculated. The treatment with Spergon always preceded inoculation. The results were the same in nonsterilized and steam-sterilized soil. The root system of peanuts in the steam-sterilized soil was larger than in the nonsterilized soil. Nodules appeared on plants in both types of soil without inoculation, but nodules on plants from noninoculated seed were larger and fewer than on roots of plants from inoculated seed. The distribution of nodules also was different.

Factors affecting the development of Melampsora lini. MISRA, A. P. *Melampsora lini* developed best at mean temperatures of 65–75° F. in the field in 1946 and 1947, but was drastically suppressed when the maximum temperature reached 92° F. or more for two or three days. Plants of Redwing and Bison flax were inoculated in the greenhouse with race 4 of *M. lini* and then exposed for 30 hours to 95–99° F. after prominent rust flecks had developed; little or no rust developed. Most urediospores collected from the field when temperatures were 98–100° F. had lost their viability and did not germinate even under optimum conditions. Partial shading of the plants in the field at 98–100° F. did not enable urediospores to retain viability; but with partial shading at 88–90° F. from 23 to 35 per cent of the urediospores remained viable. Teliospores developed well and germinated readily at 65–75° F. but not at 85° F. Ammonium nitrate fertilizer applied alone or with potash and phosphate to sandy soils greatly increased the severity of rust, whereas potash and phosphate alone did not. Teliospores collected on Koto flax at Morris, Minnesota, in August, 1946, first germinated in January, 1947. Teliospores collected on Bison at St. Paul, Minnesota, in August, 1946, did not germinate until May, 1947. Germ tubes of urediospores of race 17A grew faster than those of race 4.

Parasitic races of Ustilago tritici on spring wheats. MOORE, M. B. Nine parasitic races of *Ustilago tritici* have been found that primarily attack common wheats, and two have been found that primarily attack durums. These races were identified on the basis of the pathogenicity for 12 varieties of spring wheat, including two durums. Some varieties, such as Hope, Vernal, and *Triticum timopheevi*, are extremely resistant to all races tried; some varieties are immune from certain races and highly susceptible to others. The durums, generally resistant in nature, are very susceptible to one or both durum races when artificially inoculated. There apparently are various types of resistance, operating singly or in combinations. A Thatcher × Supriza cross and two lines from Hope × (*T. timopheevi* × Steinwedel) have been resistant when inoculated with mixtures of 4 and of 5 races, all of which were different. The variety Cadet was found to comprise two or more morphologically similar strains, one of which is resistant to several smut races that can attack the other strain.

Specificity of certain Bioquin (8-quinolinol) derivatives and some of their formulations for apple-scab control. MORGAN, OMAR D., JR., AND DWIGHT POWELL. Laboratory tests of 43 Bioquin derivatives and their formulations indicated the following as fungicides: Bioquin 1 (copper 8-quinolinolate) and its dry mixed formulations with Santocel, Celite 266, Pyrax ABB, Wyodek bentonite, Palmetto sulfur (325 mesh), and Dowmike sulfur; Bioquin 100 (zinc 8-quinolinolate) and Bioquin 300 (magnesium 8-quinolinolate) and their formulations with pyrax, bentonite, and celite. Spray tests on Golden Delicious made with Bioquin 1 (1, ¾, ½ and ¼–100), Bioquin 1 formulated with pyrax and with bentonite (25, 50 and 75 per cent Bioquin 1) at 1–100, indicated that under severe disease conditions the amount of control on the fruit obtained with the formulations of

Bioquin 1 with pyrax and with bentonite is significantly higher than when Bioquin 1 at ¾–100 is used alone. In the same test, Bioquin 100 and its formulations with pyrax and bentonite and Bioquin 300 gave slight control of apple scab. Tests on Jonathan included Bioquin 1 (1 lb.), Bioquin 1 (½ lb.) plus Factice ½ lb., Bioquin 1 (½ lb.) plus Micronized sulfur 3 lb., Puratized 1 pint, Microsul 8 lb., Phygon ½ lb., Bioquin 1 (25 per cent) plus Santocel (75 per cent) 1 lb. and Fermate 1 lb., each in 100 gallons of water. Of these, the first five gave over 95 per cent control on the fruit two months after the last application. By harvest time, the residual effect had been reduced; Puratized showed best control (72 per cent), Microsul (66 per cent), Bioquin 1 plus sulfur (63 per cent), and Bioquin 1 (62 per cent). The control of other materials ranged from 55 per cent for Bioquin 1 plus Factice to 8 per cent for Bioquin 1 plus Santocel. This last formulation gave best control in laboratory tests.

Nitrodithioacetates as potato sprays. MUNCIE, J. H., AND W. F. MOROFSKY. Potato-spray trials on the Menominee variety of potato were made by application of the nitrodithioacetates of zinc and copper and copper nitroacetate, singly and in combination, in comparison with tribasic copper sulphate and Bordeaux mixture. DDT and benzene hexachloride were added as insecticides. There was no significant difference in control of early blight. Late blight was absent. Highest yield was obtained from plots sprayed with a nitrodithioacetate of copper plus DDT. Substitution of benzine hexachloride for DDT resulted in increased infestation of potato leaf hoppers and the six-spotted leaf hoppers.

An unusual host response to certain paired monosporidial cultures of Ustilago zeae. MUNNECKE, DONALD E. Certain paired monosporidial isolates of *Ustilago zeae* produced unusual leaf curlings and distortions when hypodermically inoculated into a selfed line of corn. Stem and leaf tissues remained tightly curled, but as they grew they became gnarled, twisted, and distorted; or the leaf tip remained within the sheath and a single large loop of curled leaves resulted from the growth of basal leaf tissue. The distortions resembled immature smut galls, except that they remained green and seldom were hypertrophied. Chlamydospores never were formed, but smut mycelium was recovered when some of the distorted tissues were plated on potato-dextrose agar. In four greenhouse tests extending over six months three pairs of white cultures caused this pronounced leaf curling without resultant chlamydospore formation. The distortions were not due to mechanical injury caused by the hypodermic needle, because sterile broth check inoculations were symptomless.

Onion-mildew control in 1947. NELSON, RAY. Airplane dusting with copper-sulphur fungicides was not effective against onion downy mildew in the 1945 epidemic. Mildew did not reappear in 1946 and only yield data were obtained on effects of fungicides. Dusts and sprays were applied in 1947 to seeded Yellow Globe onions in a randomized block design with individual plots of four 30-ft rows. The first application July 7 preceded mildew detection, but all plants in the untreated plots were infected at the time of the fifth and final application on August 17. Dusts were applied at 50 lb. and sprays at 204–272 gal. per acre. DDT supplemented all materials for thrips control and Triton–1956–B as detergent in all sprays. Spray treatments were: Cuprocide; Cuprocide–p.e.p.s; Cuprocide–Dowax; Cuprocide–wettable sulphur; Dithane HE–178; Dithane HE–178–p.e.p.s; Dithane HE–178–Dowax; Bordeaux; Bordeaux–Dowax; P.e.p.s.; DDT; Wettable sulphur; Dow 606 (Zn-dimethyl-dithiocarbamate); Dow 608 (amine salt tetrachlorphenol); Dow 612 (1,4-dithio-cyano-butene); Lime-sulphur-rosin. Dusts were: Cuprocide-sulphur and Dithane HE 178. Effective mildew control with no apparent phytotoxicity was obtained with both dusts and with Dithane, Dow 612, and Bordeaux sprays. Cuprocide sprays controlled mildew but caused some injury. Significant yield increases from mildew control alone were: Dithane dust, 29 per cent; Cuprocide dust, 21 per cent; Dithane HE 178, 17 per cent; Dow 612, 12 per cent; and Bordeaux, 8 per cent.

Field trials in New York with ethylene dibromide and DD mixture against the root-knot nematode. NEWHALL, A. G., AND B. LEAR. With the aid of a tractor-drawn, three-row, sub-surface applicator, loaned and operated by the Shell Chemical Corporation, dosage rates of 200 and 300 pounds per acre of DD mixture and a 10 per cent ethylene dibromide mixture were compared for the control of root-knot nematode when applied in rows 10 and 12 inches apart in muck and upland soils. Weeds were not controlled to any extent by any treatment. Stands of carrots showed that the dosages used were near the maximum limit for spring treatments because of possible injury to seedlings and possible effect on flavor of carrots. Three tests conducted on muck soil showed a 50.9 per cent increase in yields of No. 1 carrots over checks for the DD treatments and 49.4 per cent increase for the ethylene dibromide treatments. Tests on two upland plots

showed a 155 per cent increase for DD mixture and a 122.8 per cent increase for ethylene dibromide. The data showed that the 300-lb. dosages were better than the 200-lb. dosages with both materials and application of the materials in rows 10 inches apart more effective than at the 12-inch distance.

Studies on genetics of Ceratostomella fimbriata. OLSON, E. O. Isolations from sweet potatoes affected with black rot, from six States, gave two strains of *Ceratostomella fimbriata*, the typical long-neck perithecial strain and a nonperithecial strain, the latter producing small sclerotium-like bodies. Both strains produced endoconidia, and were pathogenic to sweet potatoes. The two strains were obtained when single ascospores were isolated from the long-neck perithecial strain; this occurred for 4 generations. Single conidial cultures (164) from the long-neck perithecial strain gave only the parent type. When the two strains were paired, a line of perithecia formed where they came together. Ascospores isolated from the line gave the two strains. A third strain, with very short perithecial necks, was isolated also in one case from this line. This short-neck strain formed a line of long-neck perithecia when paired with the nonperithecial strain. Ascospore isolations from this line gave at least three strains, the long-neck, short-neck, and a nonperithecial strain. Ascospore cultures from the short-neck perithecial strain have given rise to the parental type and possibly a nonperithecial strain differing from the original one isolated from the long-neck strain.

A virus causing internal necrosis in White Rose potato. OSWALD, JOHN W. White Rose potato plants with severe leaf, stem, and tuber necrosis were observed in California in 1946. Investigation showed this to be caused by a virus easily transmissible by mechanical means. First generation symptoms consist primarily of internal necrotic browning of veins, petioles, and stems, extreme at the nodes, followed in severe cases by collapse of the entire plant. Affected leaves show irregular necrotic areas and often chlorotic blotches. As the disease progresses these may shrivel and hang. Top leaflets become rugose and tightly curled, and the leaves roll downward. Tuber necrosis occurs first just under the skin giving the surface a purplish cast. Later these areas become sunken and corky and necrosis often spreads through the entire tuber. Second generation internal necrosis is less severe. Potato seedling 41956 reacts similarly to White Rose. Free of the potato latent virus, the necrotic virus induces a chlorotic mottle in pepper, irregular yellow mottle in *Nicotiana rustica*, and green vein banding in *N. glutinosa* and *N. tabacum*. Thermal inactivation is between 50° and 55° C. and longevity *in vitro* between 48 and 60 hours. The virus properties and host reactions indicate a possible relationship to potato virus Y.

Peach X-disease on sour cherry. PALMITER, D. H., AND K. G. PARKER. Both the English Morello and Montmorency varieties of *Prunus cerasus* are affected by a virus that seems to be the same as that causing X-disease of *P. persica* and *P. virginiana*. Symptomatic plants of all three species have been found in close association in several areas of New York. The incidence of diseased sour cherry trees is usually greatest in that part of the orchard adjacent to diseased chokecherries and affected cherry trees have not been found except in association with chokecherries. Symptoms on sour cherry comprise delayed bloom, extensive June drop, especially on English Morello, and the failure of the remaining fruit to mature with full color or flavor. Internodes on the terminals may be shortened, and, during late summer and autumn, some leaves may become abnormally yellow to brick-red along their mid-veins. Graft and bud inoculations, both in the greenhouse and out-of-doors on young cherry trees, caused a wilt often quickly followed by death. Sometimes their leaves became yellow along their mid-veins. Similar symptoms on sour cherry followed the insertion of buds from X-diseased chokecherries. Inoculations from affected sour cherries to peach resulted in typical peach X-disease symptoms. In one orchard 55 per cent of the English Morello trees produced no fruit in 1947 as a result of this disease.

Apple bitter rot on sour cherry in West Virginia. PEET, CLYDE E., AND CARLTON F. TAYLOR. A rot of sour cherry, previously unreported as an economic disease in North America, occurred in appreciable amounts in two orchards and in small amounts in the other orchards examined during the unusually wet 1947 harvest season. The incidence of the rot varied considerably from tree to tree. On some trees 15 per cent (by count) of the fruits were infected, with all fruits attacked on some branches. The rot first appeared as a small tan spot, then progressed rapidly to affect approximately half the fruit area. The surface of the lesion became darker, tended to be folded concentrically, and Gloeosporium-like spores were produced in pinkish masses. Successful cross-inoculation was obtained on Grimes Golden fruits when infected cherries were suspended in cheesecloth bags in the tree a few inches above the apple or when an opened

Petri-dish culture of the cherry isolate was suspended similarly. Lesions typical of apple bitter rot resulted. Growth characteristics of isolates from affected cherries were similar to those of isolates from typical apple bitter-rot lesions. Preliminary studies and the successful inoculation of apple suggest that the rot on cherry is caused by *Glomerella cingulata*.

Resistance to powdery mildew in peas. PIERCE, W. H. In repeated field tests a selection out of the Stratagem variety of garden peas has shown a high degree of resistance to powdery mildew, *Erysiphe polygoni*. Since powdery mildew is often severe in the arid sections of the western States, particularly on late home-garden varieties, a few crosses were made using the resistant Stratagem selection with the standard varieties Glacier and Shasta. In F_2 populations of 330 plants, counts of resistant and susceptible plants gave 243 susceptible and 87 resistant. The total F_2 population (139 plants) of the Stratagem-Shasta cross was tested again in F_3. The plants classified as resistant in F_2 gave F_3 progeny which were also resistant, while plants classified as susceptible in F_2 gave progeny one-third of which were completely susceptible and two-thirds segregating in the approximate ratio of 3 susceptible to 1 resistant. Inheritance appears to be on a single factor basis with resistance recessive. Acceptable horticultural types have been selected from these hybrid progenies and are being increased for introduction.

Types of scab lesions on Camellia. PLAKIDAS, A. G. Necrotic lesions of various types occur on camellia leaves in Louisiana. The following types are recognized. Type I: General brownish discoloration of the leaf and irregular black spots occurring usually near the margins or irregularly scattered on the upper surface. Type II: Circular to irregular snowy-white, slightly raised, spots on the upper surface. Type III: Raised, brown to blackish, angular spots on the upper surface, 1–20 mm. with prominent angular cracks through the center, and, in the larger spots, with fine concentric cracks. Type IV: Spots on the upper surface, large, typically circular, brown with grayish centers, raised, with very numerous concentric cracks. Type V: Spots typically on the under surface, rarely on the upper. Lesions start as water-soaked pustules, later erupting to form brown, hard, corky excrescences, in small groups, or covering most of the leaf surface. Type VI: Small, circular to irregular, slightly raised, white to light gray, spots on the upper surface. This type occurs most commonly on *C. sasanqua*: the other types are more common on *C. japonica*. The nature of the lesions on camellia was unknown until 1942, when S. C. Arruda isolated *Sphaceloma* sp. from types I, II, and VI. Since then, the writer has repeatedly isolated *Sphaceloma* from lesions of types I, III, IV, and V. It has not been definitely determined if more than one species of *Sphaceloma* is involved.

Cabbage blackleg from seed grown in Puget Sound. POUND, GLENN S., AND O. H. CALVERT. For more than twenty years the Puget Sound area of western Washington has been this nation's chief producer of cabbage seed, primarily because seed grown there has not been known to carry the blackleg (*Phoma lingam*) and blackrot (*Xanthomonas campestris*) organisms. Such seed has been used commonly without hot-water treatment because of this known freedom from infection, although a mildly pathogenic strain of *P. lingam* has been recorded recently from that area. In 1947 a severe epidemic of blackleg occurred in most cabbage-producing sections of the country. Extensive field surveys indicated transmission from Puget Sound seed. Isolations have been made from several seed lots grown in the Puget Sound area in 1946 and *P. lingam* has been repeatedly isolated from 4 lots, representing a growing area 200 miles in length. Koch's postulates have been repeatedly applied to isolates obtained, establishing proof of blackleg transmission on Puget Sound seed. Certain isolates obtained from seed show cultural and pathogenic characteristics of the mild strain described from the Puget Sound area while the other isolates are typical of virulent eastern strains. This suggests a probable increase in pathogenicity of the Puget Sound strain and renders use of seed grown there without hot-water treatment unreliable.

Strains of cucumber mosaic virus pathogenic on crucifers. POUND, GLENN S., AND J. C. WALKER. A mosaic disease of the ornamental crucifer, dame's violet (*Hesperis matronalis*), in Wisconsin has been found to be due to two closely related strains of *Cucumber virus 1*. Both viruses were widely pathogenic on members of the Cruciferae including the cabbage tribe. No prominent symptoms, however, were produced on cultivated species tested other than dame's violet. The physical properties of the two viruses agreed very closely with those of *Cucumber virus 1*. The points of inactivation were: tolerance to dilution, 1 to 100,000; aging *in vitro*, 4 to 5 days at 20° C.; thermal inactivation, 70° C. for 10 minutes. One of the two viruses was shown to effectively immunize zinnia (*Zinnia elegans*) plants against Price's no. 6 strain of *Cucumber virus 1*.

Anthracnose of peaches on the market. RAMSEY, G. B., M. A. SMITH, AND B. C. HEIBERG. During July, 1947, Georgia peaches arriving on the northern market showed an unsual type of decay never before observed in commercial shipments. In some carloads up to 12 per cent of this rot was found in various stages of development. Brown lesions 1/4 to 3/4 inches in diameter were most common. The larger lesions bore acervuli with salmon-pink spores characteristic of the anthracnoses. Numerous isolations from typical lesions always yielded pure cultures of *Glomerella cingulata.* Inoculation experiments showed the organism to be pathogenic to peaches, apples, pears, and plums without evident wounds as well as through wounds. Commercially brushed firm-ripe peaches in contact with mycelium or conidia in a water suspension developed lesions 16 mm. in diameter within a week at 85° F. Under similar conditions inoculations through wounds produced lesions 36 mm. in diameter. No visible infections were obtained within a week when wounded and unwounded peaches were inoculated and held at 45° F.

Metal reagents as antisporulants. RICH, SAUL, AND JAMES G. HORSFALL. The prevention of fungus sporulation has been suggested as a possible method of minimizing the loss from phytopathogenic fungi. As fungus spore production has been shown to be related to heavy metal metabolism, compounds which combine readily with metals were tested *in vitro* as antisporulants. The metal-reacting oximes and quinolines were found to be effective in reducing the spore production of *Sclerotinia fructicola, Penicillium* sp., *Alternaria* sp., and *Aspergillus niger.* Dimethylglyoxime, which forms chelate salts with most of the heavy metals, reduced the sporulation of *Aspergillus niger* growing on complete liquid media. Increasing concentrations of dimethylglyoxime decreased sporulation to the point of complete inhibition of spore production without appreciable growth reduction. Yarwood (1941) found that H_2S inhibited the sporulation of *Peronospora humuli.* Care was exercised in this study to distinguish between fungistasis and antisporulation.

Pathogenicity of isolates of corn root-rot organisms in Mexico. ROBLES, LEONEL H. Among 146 isolates of the corn root-rot organisms from 8 States, *Helminthosporium sativum* was the most destructive, while species of *Fusarium* were found to be most prevalent but much less active as root-rot pathogens. In preliminary pathogenicity test four isolates were selected: one of *H. sativum,* two of *Rhizoctonia* spp. and one of *Diplodia zeae.* When these organisms were tested with 26 different types of corn, only *H. sativum* proved to be strongly pathogenic. When all possible combinations among the four isolates were made the mixture of *H. sativum* and *Rhizoctonia* spp. was moderately pathogenic in contrast with *H. sativum* alone. The general result of this combination was a considerable reduction in the percentage of seed germination.

Barley scald in Mexico. RODRÍGUEZ VALLEJO, JOSÉ. In 1947 barley scald caused by *Rhynchosporium secalis* was reported from Mexico for the first time. The disease was found attacking barley in the experimental plots at the agricultural experiment station in Chapingo, Mexico. Apparently the disease is not widespread and thus far has been found only at the station in Chapingo. Those introduced varieties exhibiting greatest susceptibility are: Club Mariout, Atlas, Velvet, Peatland, Chevron, Kindred, Hanna, Wisconsin Barbless, and Bolivia × Chevron while Mars, Tregal, Manchuria, Bolivia, Svansota, and Texan (CI–5127) were not infected this year. The so-called native barleys planted at the experiment station ranged in reaction from highly susceptible to fairly resistant.

Specificity of fungicides for diseases of bent turf. ROWELL, JOHN B. Five years of investigation have repeatedly demonstrated that no single chemical will completely control all of the following bent turf diseases: dollar spot (*Sclerotinia homeocarpa*), pink patch (*Corticium fuciforme*), copper spot (*Gloeocercospora sorghi*), large brown patch (*Pellicularia filamentosa*) and helminthosporium blight (*Helminthosporium* sp.). Newly developed organic cadmium and mercury compounds have proved to be more effective than fungicides used previously. Cadmium compounds have shown the least specificity; five organic and two inorganic forms control the first two diseases, and three of these organic forms also control copper spot. Five phenyl mercurials compared in 1947 varied in degree of disease control and injury to bent grasses (*Agrostis* spp.) and Crabgrass (*Digitaria sanquinalis*). To date none of the compounds has given significent control of large brown patch or helminthosporium blight. These investigations indicate that a fungicide program may be necessary for maintaining healthy turf.

Root-rot of the Mexican "frijol." RUPERT, J., B. ORTEGA, AND C. CARDONA. During the past three years studies have been in progress with regard to root-rot of the frijol (*Phaseolus vulgaris*), in Mexico. Tremendous variation has been observed within

the varieties commonly grown in Mexico with regard to resistance to this disease, but, with few exceptions, the Mexican bean is susceptible. Two organisms appear to be primarily responsible for root rot of beans in Mexico, and these are *Sclerotium rolfsii* and *Rhizoctonia solani*, although various species of *Fusarium* are frequently isolated and may be involved in this complex. When bean seedlings are inoculated with isolates of *Rhizoctonia* or *Sclerotium*, infection results and typical root-rot symptoms develop. Selections are being made in an attempt to secure resistant lines for a breeding program in 1948.

Development of Cercospora-resistant strains of rice. RYKER, T. C., AND L. E. COWART. To secure varieties of rice resistant to the Cercospora leaf spot (*Cercospora oryzae*) consideration must be given to the parasitism of the different races of the fungus as well as to the resistance of the host. Eight races of the fungus based on their ability to attack 7 test varieties, Blue Rose, Blue Rose 41, Caloro, Fortuna, Red Rice, Rexoro, and Nira have been reported. Two additional races have been identified, race 9, to which Shoemed and three of the test varieties, Blue Rose, Fortuna, and Nira, are susceptible, and race 10, which is similar to race 2 except that it attacks Caloro as well as Blue Rose and Blue Rose 41. Inheritance studies in crosses involving race 6 (Rexoro strain) indicated a single dominant factor for resistance in 5 crosses and two dominant factors for resistance in 3 crosses. All of the commercial varieties used in the rice breeding program are susceptible to one or more parasitic races of the fungus. Delrex, a recently released selection from a cross between Rexoro and Delitus and a number of advanced selections from crosses between Rexoro and Blue Rose are resistant to all of the known parasitic races of the fungus. Thus the factors for resistance in rice to *Cercospora oryzae* may be combined through hybridization.

Chemicals for control of the golden nematode of potatoes. SCHMITT, C. G. Allyl bromide, D-D trimethylene bromide, ethylene bromide, and Iscobrome D were all rather effective in kill of cysts of the golden nematode in a pot test at 23 gal. per acre. Of the chemicals used, the only ones which eradicated under these ideal conditions at 76 gal. per acre were ethylene dibromide and Iscobrome D.

Resistance to apple scab in certain clones of Malus species. SHAY, J. R., AND L. F. HOUGH. Twenty-two clones of *Malus* in plots at Urbana, Illinois, and in Morton Arboretum, Lisle, Illinois, have remained free from foliage and fruit infection by *Venturia inaequalis* during the past three years in which apple scab has been epiphytotic. These clones were grown in the greenhouse where controlled inoculations were conducted. Heavy conidial suspensions from 15 single-ascospore cultures of *V. inaequalis* isolated from 5 apple varieties at Lafayette, Indiana, were combined in each of 4 inoculation series and used singly in 2 inoculation series. On the basis of maximum symptom development, the clones could be divided into four groups: 1. No evidence of infection (*M. toringo* and *M. ioensis*). 2. Numerous or few pin-point depressions with no sporulation (*M. micromalus*, *M. floribunda*, and 11 other clones). 3. Irregular, necrotic or chlorotic lesions varying from 2 to 5 mm. with no sporulation (*M. baccata jackii*, *M. sikkimensis*, and a Russian seedling). 4. Lesions few, elongate or irregular, necrotic or chlorotic; sporulation when present sparse (*M. baccata* and 3 other clones). One isolate was unable to produce macroscopic symptoms on *M. sikkimensis*. Heterozygosity and dominance of genes controlling resistance of 8 clones were demonstrated by inoculation tests of seedling, F_1 progenies of these clones crossed with susceptible varieties.

The brown-spot disease of celery. SMITH, M. A., AND G. B. RAMSEY. A serious field and market disease of celery from Colorado not previously reported in the United States has been studied since 1943. The disease is characterized by brown, discolored areas in the form of circular to oblong lesions 1/16 to 3/4 inches in diameter on the inside surfaces of the stalks. On the outer stalk surfaces the lesions are somewhat smaller and more elongated. Infection also occurs on stalklets, in leaflet axils, and on leaflet blades. A *Cephalosporium* sp. constantly associated with infected stalks, stalklets, and leaves on commercial celery bunches has been isolated many times. Typical infections have resulted from pure culture inoculations of celery bunches as well as celery seedlings grown in the greenhouse. The organism has been reisolated in pure culture from lesions resulting from these inoculations. In potato-dextrose agar plate cultures the optimum temperature for growth was 75° F., while growth was not observed at the two extreme temperatures 40° and 90°. Optimum growth of the organism occurred at pH 6.5. There was no growth at pH 4.0.

Classification and identification in Fusarium. SNYDER, W. C., AND H. N. HANSEN. Great variability between clones of wild types, and between their derivatives, is common

throughout the genus *Fusarium*. A classification is required that puts together into one species all such entities of that species, whether they represent clones existing in nature or derived in culture, in spite of any morphologic, biologic or apparent differences resulting from mutation, environment, or host relationships. Such a system would recognize few species, based on morphology, indicate pathogenicity by *forma* names and races within *formae* by numerals, and has been found in practice to facilitate identification. Spore size has been used extensively in the past for speciation, yet spore dimensions of wild types of a fungus vary so greatly as to fall into several of the older species. Consistent differences in spore size do not establish a difference in species because such differences can be shown among single-spore isolates from a given clone. Also, the older concept of basing species on hosts falls apart when required to handle the various races that Armstrong has shown are pathogenic on several different combinations of hosts.

The relation of barberries to physiologic races of Puccinia graminis in the United States in recent years. STAKMAN, E. C., AND W. Q. LOEGERING. With the progress of barberry eradication in the United States there has been a general trend toward stabilization in the prevalence and distribution of races of *Puccinia graminis tritici*. In 1945 and 1946 only four races—17, 19, 38, and 56—were prevalent and widespread in the country as a whole. In the two years, races other than these comprised only 5 and 3 per cent of the isolates identified in areas in which few barberries remain. In barberry-infested areas of Pennsylvania, Virginia, West Virginia, and Washington, on the other hand, new or unusual races or biotypes comprised 26 and 24 per cent of the total number of isolates. The same general relationship appears to hold for 1947. None of the new races or biotypes so far found has a wider host range than race 15B. With one exception, race 7 of *P. graminis avenae*, which attacks White Tartar oats and varieties that derived stem rust resistance from it, has been found only near rusted barberries in Northeastern United States, where it attacked certain normally resistant varieties very heavily in experimental plantings near barberries. Elsewhere, only races 2, 5, 8, and 10 have been found in recent years.

Infection of the sugarcane stalk by red rot. STEIB, R. J. When stalks of sugarcane, which appeared healthy, were surface-sterilized up to 24 hours with a solution of bichloride of mercury and cultured, the red-rot organism (*Physalospora tucumanensis*) was obtained from root bands and buds. It was isolated also from leaf scars and small red areas found in the tissues under the leaf scars. The organism was not obtained from nodes enclosed by tight, healthy leaf sheaths. Several sugarcane varieties developed red rot, even after fungicidal treatments and subsequent storage with the fungicide residue remaining on the cane; the buds and leaf scars served as the initial points of entry. Various treatments with fungicides and hot water did not eliminate the organism from the stalk. Leaf sheaths of susceptible and resistant varieties collected in the field developed red rot when placed in moist chambers. The perithecial stage occurred on a large percentage of dried leaf sheaths of Co. 290. The evidence indicates that the sugarcane stalk is infected with the red rot organism previous to planting, perhaps in a dormant form, and that the disease develops when or if the vitality of the cane becomes lowered.

Time and frequency of occurrence of Pythium debaryanum and graminicolum on roots of barley. SUMMERS, THOMAS E. Seven field plantings of barley were made from April 2 to July 9. *Pythium debaryanum* was frequently isolated from germinating seeds, roots of young seedlings, and roots of older plants from April 7 through the first week of July. After that time it was isolated with decreasing frequency until July 24 when none was recovered. *P. graminicolum* was not isolated from germinating seeds or from roots of very young seedlings. However, a high percentage of isolations from roots of older plants in all plantings contained *P. graminicolum*. The fungus was recovered frequently from May 2 through August 3. Among isolations made from roots of plants less than eleven days old only 5.6 per cent contained *P. graminicolum*. Among isolations from roots of plants more than eleven days old, 73.5 per cent contained *P. graminicolum*. In no case was *P. graminicolum* isolated from germinating seeds; it was rarely isolated from roots which had not attained a length of 8 cm. *P. debaryanum* was frequently isolated from germinating seeds, roots of very young seedlings and roots of older plants grown in field soil in the greenhouse. *P. graminicolum* was recovered only from roots of older greenhouse-grown plants. Apparently *P. graminicolum* does not infect the germinating seed or roots of very small barley seedlings in field soil.

The effect of cereal crop rotation on "take-all" damage. SUNESON, COIT A., AND JOHN W. OSWALD. A randomized series of wheat, barley, oat, and rye plots grown for hay yield trials in 1946 disclosed severe "take-all" damage in the first two mentioned

crops. The absence of separating alleys and minimum intervening tillage facilitated replanting the same crops in 1947 at right-angles to the previous seeding. From the resulting checkerboard, yields of each crop succeeding itself and each of the other crops, expressed as grams of grain and straw per square foot, were determined from averaging yields at six points. Thus, on areas previously cropped to wheat, the current yields of wheat, barley, oats, and rye were 8, 29, 51 and 55 grams, respectively. On previous barley land these crops yielded 8, 30, 54, and 58 grams; on previous oats, 42, 52, 42, and 51 grams; and on previous rye, 45, 57, 48, and 54 grams, respectively. All isolations from both years' croppings of wheat and barley yielded *Ophiobolus graminis*. No pathogenic fungi could be isolated from oats or rye in either year. The data clearly depict the relative susceptibility of the four crops, their effect on the survival of *O. graminis*, and the benefit from rotating rye or oats with barley or wheat. (Cooperative investigations, Bureau of Plant Industry, Soils, and Agricultural Engineering, U. S. Department of Agriculture and California Agricultural Experiment Station.)

Barley powdery mildew (Erysiphe graminis hordei) influenced by environment previous to inoculation. TAPKE, V. F. Wide differences of opinion have long existed in regard to the environmental conditions that promote powdery mildew in barley and wheat. In a preliminary study of environmental influences, two susceptible varieties of spring barley were sown in pots in September, 1946. Soon after seedling emergence some of the pots were put outdoors, some were kept in a greenhouse and lightly watered, and some were kept in a greenhouse and liberally watered. After 8½ weeks the plants grown under these different conditions were mildew-free. They were then uniformly inoculated with powdery mildew conidia and thereafter amply watered and kept in a greenhouse under similar conditions. On the outdoor plants, which were short and heavily tillered, very little mildew developed; on the lightly watered greenhouse plants, a moderate amount developed; and on the heavily watered greenhouse plants, which were tall and succulent, there was very abundant mildew. These results indicate that the pre-inoculation environment may materially affect the post-inoculation reaction of the plants to mildew. This subtle pre-inoculation influence, hitherto unsuspected, apparently, may explain the divergent reports on post-inoculation conditions that promote powdery mildew.

Prolonging viability of spores and mycelium of the barley loose smut, Ustilago nuda. TAPKE, V. F. Spores of many different collections of *U. nuda* and mycelium in seed from hand-inoculated flowers were stored at 32° F. in a refrigerator. At these temperatures some of the spores were still viable after nine years, whereas in storage at room temperatures, viability often declines appreciably in three to six months or less and seldom exceeds one year. Seeds of five barley varieties from flowers inoculated by hand in 1940 and stored at 32° F. were sown in a greenhouse in January, 1947. Sixty-seven per cent of the seeds produced seedlings. In the variety Lion (C.I. 923), 73 per cent of the heads were smutted. Knowledge of the possibilities of cold storage for prolonging viability in *U. nuda* has facilitated study of its physiologic races and breeding for resistance.

Bacteria in healthy plant tissue. TERVET, IAN W., AND JOHN P. HOLLIS. Potato plugs obtained aseptically from many apparently healthy tubers were found to have bacteria associated with their tissues. Plugs prepared with care to avoid superficial or aerial contamination, when placed in tubes of sterile water, not infrequently disintegrated and deposited starch. Besides complete breakdown, 4 additional types of bacterial action were evident: turbidity with gas; green fluorescence; pellicle formation; and partial breakdown without starch liberation. From 15 lots of sound Red Warba potatoes, over 1000 plugs were made and submerged in sterile water. Bacterial development ranged from 10 per cent of the plugs in some lots to about 90 per cent in other lots. Plugs obtained from aerial tubers also were frequently found to contain bacteria. Similarly prepared plugs from storage organs of several plants developed thus: 80 per cent of the turnips gave bacteria, about 30 per cent for carrots, beets, and kohlrabi, while sweet potatoes and tomato fruits remained sterile. Variation in disinfecting procedures and in methods of obtaining plugs did not vary appreciably the percentage of plugs with bacteria. The results indicate that at least some apparently healthy, fleshy storage organs may harbor bacteria.

Germination of spores and artificial culture of Urocystis agropyri from red top. THIRUMALACHAR, M. J., AND J. G. DICKSON. *Urocystis agropyri* occurs on many important pasture grasses. Recently, Fischer suggested the combination of *U. tritici* and *U. occulta* under *U. agropyri*. Germination of the chlamydospores of *U. agropyri* from any of the grass hosts so far is unreported. Chlamydospores on *Agrostis gigantea*

collected at Madison were germinated and the cycle of the fungus studied. A whorl of 2 to 4 sporidia are borne at the apex of the promycelium; these fuse in pairs and develop infection hyphae. Occasionally two or more nuclei pass directly from the promycelium into the sporidia, resulting in infection hyphae without sporidial fusions, as described by Stakman *et al.* in *U. occulta*. Cultures of the fungus isolated have been strictly mycelial types, although cultures from haploid sporidia have not been tested. Chlamydospores developed in culture consisted of spore balls without apparent sterile cells.

The effects of representative plant growth substances upon attenuated-bacterial crown galls. THOMAS, JOHN E., AND A. J. RIKER. Plants inoculated with an attenuated (A6–6) strain of the crown-gall bacteria (*Phytomonas tumefaciens*) have been treated with 57 different, representative plant growth substances. The plants were decapitated and the chemicals in lanoline applied to the cut about 2 inches above the bacterial puncture inoculations. Results were secured from treatments of more than 4900 plants, including tomato, sunflower, Paris daisy, marigold, and velvet leaf. Such plants treated with 19 of these substances developed attenuated-bacterial galls which were larger than those on the controls. These 19 substances represented numerous different kinds of plant growth substances. Attenuated-bacterial-gall stimulation by these chemicals was sometimes, but not always, associated with such growth responses as epinasty, root stimulation, bud suppression, tissue proliferation, or formative effects. Consequently the increased growth of the treated attenuated-bacterial galls may be considered as another type of growth response. Bacteria, reisolated from the stimulated galls were still attenuated. The effect of the growth substances in stimulating the attenuated-bacterial galls appeared to be on the host, or the bacteria-host interaction, rather than on the bacteria.

Differential tests of three isolates of Phytophthora infestans. THOMAS, W. D., JR. One isolate of *Phytophthora infestans* from Maine and 2 from Colorado were tested for their pathogenicity on 10 potato selections and 5 tomato varieties. Of the latter isolates, 1 was from Weld County and 1 from the San Luis Valley. The Weld County isolate was least pathogenic on potatoes, and the Maine isolate was least pathogenic on tomatoes. The Red McClure potato and Pritchard tomato were moderately resistant to foliage infection by all isolates. Simultaneous studies on the retention of sporangial viability in different relative humidities at 24° C. showed that the San Luis Valley isolate survived at a relative humidity of 25 per cent for 3 hours, the Weld County isolate 1 hour at 35 per cent, and the Maine isolate 1 hour at 45 per cent. Some sporangia of the San Luis Valley isolate survived 48 hours in a 55 per cent relative humidity. After 5 hours, 85 per cent relative humidity reduced significantly germination by the Colorado isolates, and 95 per cent reduced the Maine isolate. This evidence indicates that the isolates are separate strains of the pathogen, the Colorado forms capable of surviving conditions of lower atmospheric moisture.

The rôle of minerals in production of streptomycin by Streptomyces griseus. THORNBERRY, H. H. Some plant and animal products that stimulate the production of streptomycin when added to a basal medium (glucose 10 gm., peptone 5 gm., NaCl 5 gm., distilled water 1 liter) were ashed at 700° C. and added to the same medium to ascertain whether the organic or mineral constituents of these products were responsible for streptomycin production. Stimulation expressed as the ratio of production to that in the basal medium (15 units per ml. in 3 days of shaken culture at 26° C.) was obtained: from corn steep 12.9, cowpea seed 12.1, distiller's dry solubles 24.8, peanut meal 9.3, peanut protein 3.4, peanut aqueous extract at pH 8.0 11.5, peanut protein waste liquor 18.5, peptone 6.0, sugar-beet hulls 14.6, sunflower seed meal 23.8, and yeast extract 9.3. There was a reduction in streptomycin production by the ash of some materials: alfalfa meal 0, apple leaves 0.46, chestnut leaves 0.73, and wood sulfite waste 0.93. Peanut meal contained some organic inhibitor since all ashed fractions gave higher production than non-ashed fractions. From this evidence it is concluded that minerals play an important rôle in streptomycin production.

Studies on Xanthomonas pruni bacteriophage. THORNBERRY, H. H., A. EISENSTARK, AND H. W. ANDERSON. *Xanthomonas pruni* bacteriophage has been observed to remain active and specific for its host for 20 years at room temperature, in a sealed glass tube and away from light. Lysis occurred in the first inoculation in which 0.1 ml. of the stored phage was added to a 5-ml. broth culture. The titer of the phage preparation when stored was 10^{-5}. It was not determined when revived. By serial passage through broth cultures with heavy growth (24-hour growth in broth with 1.8 cm.^{2} air surface per ml.), the titer was 10^{-9} on the 12th serial transfer. This preparation of phage lysed all cultures of *X. pruni* that were isolated from peach, plum, and apricot from various parts

of Illinois. It did not lyse *X. lactucae-scariolae,* which is distinguishable from *X. pruni* only by pathogenicity, or *Erwinia amylovora* isolated from apricot twigs, or *X. campestris* var. *armoraciae.* Electron micrographs of the bacteriophage show that it is similar in size and form to *Escherichia coli* bacteriophage and that the bacteriophage is absorbed by *X. pruni.*

Chromates as potato fungicides. THURSTON, H. W., JR., J. G. LEACH, AND J. D. WILSON. Since 1941 numerous compounds containing chromium together with from one to four other metals have been under test, as potato fungicides in Pennsylvania. During 1946 and 1947 certain of the most effective of these have been tested as well in West Virginia, Ohio, and Florida. Certain copper zinc chromates have given outstanding results in the control of both early blight (*Alternaria*) and late blight (*Phytophthora*). When these diseases are serious enough and infection takes place early enough commensurate increases in yield are noted. When used alone, these chromates have been shown to have insect-repellent properties equivalent to Bordeaux mixture and when used with DDT the yields produced have equaled or surpassed the yields from "fixed coppers," "Dithane," and Bordeaux.

An inoculation method for determining resistance in corn to Diplodia ear rot. ULLSTRUP, ARNOLD J. Inoculum was prepared from spores produced on sterile whole oats in 2-qt. fruit jars after 4 to 8 weeks of incubation in the light at 22° to 26° C. Spores from 2 jars of culture were washed free of the substrate and suspended in 1 qt. of water. The final inoculum was made up of 1 qt. of stock spore suspension in 2.5 gal. of water. The inoculum was sprayed over the corn ears by means of an ordinary compressed-air sprayer. Approximately 300 ears could be sprayed with 2.5 gal. of inoculum. The optimum time for inoculation appeared to be a period from full silk until 2 to 3 weeks later. Inoculations made 4 or 5 weeks after full silk resulted in much lower infection. No differential virulence was observed in several isolates tested. Isolates vary considerably in sporulating ability. Selection of isolates was made on the basis of rapid and abundant spore production as determined by preliminary tests. Incidence of infection was determined by (1) percentage of infected ears, (2) by percentage of infected kernels, by weight, in a shelled aliquot of each plot, and (3) by a disease index. Results of the three methods showed a highly significant correlation. Resistance of inbred lines was determined in single-cross combination.

Antibiotics and potato ring rot. VAN SCHAACK, VALERIA. Penicillin and streptomycin kill the potato ring-rot bacterium (*Corynebacterium sepedonicum*) *in vitro.* Therefore, they were applied to artificially inoculated, cut seed pieces. Potatoes used were Minnesota-grown, certified Pontiac and White Rose varieties. Inoculated, but antibiotically untreated seed-pieces of neither variety grew, but rotted in the soil; there was good growth of controls. Inoculated and penicillin-treated seed-pieces did not grow, although controls produced healthy plants. Inoculated, streptomycin-treated seed-pieces gave good growth with both potato varieties.

Combining resistance to wildfire, mosaic, black root rot, and fusarium wilt in burley tobacco. VALLEAU, W. D. In 1946 Ky 23 burley tobacco was pollinated with *Nicotiana longiflora* pollen, a species resistant to wildfire. Several pods were well filled but nearly all seeds were hollow. The forty fertile seeds obtained were treated with colchicine during germination. Of 31 plants, four proved to be fertile when selfed or backcrossed. Resistance to wildfire is dominant, so each generation was backcrossed to eliminate, as quickly as possible, all *N. longiflora* chromosomes except the one, or a portion of it, containing the resistance factor. The backcross parents used were varieties resistant to mosaic (NN) and black root rot; or to mosaic (NN), black root rot, and fusarium wilt. The first and a few of the second backcross generation plants were strap-leaved and rosette-like, but others of the second backcross were excellent type burley plants. The third backcross generation is all tobacco-like. It is hoped that by this method of breeding, varieties will soon be developed that are practically immune to four major tobacco diseases. These varieties of *N. tabacum* will contain genes from *N. longiflora* and *N. glutinosa.*

Factors affecting the nature of resistance of potatoes to scab. VAUGHN, JOHN R. Certain pustule types of scab were first noticed to differ from the common scab type among seedlings tested at the Minnesota Agricultural Experiment Station in 1938. Experiments in the field and greenhouse were conducted for three years at Minnesota and later for one year at West Virginia. These experiments showed that two of the abnormal pustule types selected for study were constantly associated with certain seedling lines, regardless of the race of the pathogen causing infection. This indicated that

the expression of resistance or susceptibility in these cases depended upon the genetic constitution of the host. The Menominee variety, which has consistently been resistant to the attack of many races in different locations in the United States, was investigated to determine the mechanism of its resistance. It was found that the wound periderm formation was completed three times as fast in this variety as in the susceptible Smooth Rural variety. It is suggested that the type of resistance found in Menominee is due to the speed of wound periderm formation, which results in the exclusion of the pathogen from extensive penetration of the host tissue.

Development of clubroot resistance in cabbage. WALKER, J. C., AND R. H. LARSON. Long search for individuals in cabbage varieties, that were resistant to clubroot (*Plasmodiophora brassicae*), has been unsuccessful. In 1941 kale-cabbage hybrid rogues were found in a Wisconsin cabbage field, self progenies of which contained highly resistant individuals. Some of the fourth-generation inbred progenies have uniformly high resistance. These inbreds have the leafy headless kale character. In crosses with cabbage the F_1 generation segregated with a preponderance of susceptible individuals. In the F_2 a small percentage of highly resistant individuals appeared, which were divided about equally between kale, intermediate, and heading plants. From the last group, selections have been made to secure lines in which high resistance is combined with desired head and plant type.

Multiple alleles and factor interaction in Glomerella. WHEELER, H. E., AND S. J. P. CHILTON. Single ascospores from plus-type cultures of *Glomerella* from a variety of host plants invariably produced a high percentage of variants of the minus type. Less frequently variants of other types were produced. During a six-year period a total of eighteen distinct strains, differing in various cultural and morphological characteristics, have been isolated from the progenies of a plus culture from wild morning glory. Analyses of asci produced by appropriate crosses indicated that the differences between seven of these strains were controlled by six genetic factors or groups of factors. Three of these factors were at one locus (A) and three at a second locus (B) on the same chromosome. The factors at each of these loci apparently represent multiple alleles. Two pairs of allelomorphic factors, one at each of the two loci, conditioned the production of fertile perithecia. A factor for partial self-sterility at locus B was epistatic to a factor for self-fertility at locus A when the two were combined in a strain of the minus type. The reverse was true when the same two factors were combined in a strain of the plus type.

Gomphrena globosa, a useful plant for qualitative and quantitative work with potato virus X. WILKINSON, R. E., AND F. M. BLODGETT. Local lesions, without systemic infection, develop when *Gomphrena globosa* the globe amaranth, is inoculated mechanically with potato virus X, provided carborundum is used. Many strains including so-called masked, mottle, and ring-spot strains were tested and all gave this reaction. Lesions appear in 2 to 3 days and are encircled by a red ring within a week after inoculation. Lesions form more slowly and less distinctly on old leaves than on young leaves. Although plants tested from different lots of seed varied considerably in size and appearance, they all appeared to react in the same manner except that some produced many more lesions than others when inoculated with a given inoculum. Since this plant has opposite leaves, which are similar in appearance and sensitivity, it is well-adapted for quantitative work. Most satisfactory results were obtained when plants were used just prior to flowering. At this stage, most plants have 4 pairs of leaves which are large enough for quantitative work. The number of lesions formed is increased if the leaves are rinsed immediately after inoculation, and is decreased if the leaves are shaded for 48 hours before or after inoculation.

A detached leaf method for vector studies with the tomato-spotted-wilt virus. WRIGHT, C. M., AND F. M. BLODGETT. The onion thrips, *Thrips tabaci,* was successfully reared on potato or tomato leaflets placed in Petri dishes containing water. Nymphs were confined on the detached leaflets by a ring of Tanglefoot. Emerging adults were placed in microcages mounted on leaflets. Some adults that had previously fed as nymphs on spotted-wilt-infected potato leaflets produced local lesions when they were confined on healthy detached petunia leaves. These lesions appeared three days after the infective adults were allowed to feed on the petunia leaves, and were similar in appearance to lesions resulting from mechanical inoculation with the virus. The study indicated the importance of maintaining pedigreed cultures of thrips, of testing for the presence of the virus in the leaflets on which the nymphs were to be confined, and of mechanically inoculating one-half of the petunia leaf with the virus to make certain that it reacts with local lesion formation. Leaves of *Nicotiana glutinosa* and *N. tabacum* could not be used to test the infectivity of the adults because the insects failed to feed when confined on them.

Hereditary defects in tomatoes. YOUNG, P. A. Inherited defects in tomatoes resemble viroses or other plant diseases. For example, inherited necrosis caused epinasty of rhachises, browning of veins, yellow spots in leaflets, dying of leaflets, and browning of the epidermis of tomato stems. Inherited lutescence caused yellowing and browning of the lower leaves and yellowing of the stems and unripe fruits. Pits and mosaic-like stripes were inherited in the fruit peel. Brachytic and extremely dwarfed types of growth resembled the bunchy-top viroses. Runty plants remained dwarfed in cold frames. Wiry leaves and midget tomatoes resembled witches' broom or extreme fern leaf. Wilting leaves suggested a root disease. Yellow lethal seedlings died early. Sticky-gene hybrid tomatoes were sterile when they resembled their *Lycopersicum chilense* parent. Inherited susceptibility to physiologic abnormalities occurred in tomato varieties with high percentages of leaf roll, fruit puffing, blossom-end knobs, cuticle cracks, blossom-end rot and catface. A mutant tomato had extremely puffed fruits resembling bell peppers. When genes for resistance to parasitic diseases exist, their alleles for susceptibility are inherited defects, such as susceptibility to wilts caused by *Fusarium* and *Verticillium*, and to leaf spots caused by *Cladosporium* and *Septoria*.

Pod mottle, a virus disease of beans. ZAUMEYER, W. J., AND H. REX THOMAS. A new bean virus, which produces local lesions in certain varieties and systemic infection in others, was isolated in 1945 from mottled bean (*Phaseolus vulgaris*) pods collected in South Carolina. The local lesions caused by this virus are different from those caused by any other virus affecting beans. The systemic mottle symptoms resemble those produced by southern bean mosaic virus but are more intense. Pod mottle virus produces a decided mottling and considerable pod malformation The virus is readily transmitted mechanically. Immunological studies showed no relationship between pod mottle, southern bean mosaic, and common bean mosaic viruses. Just as in the case of southern bean mosaic, a variety susceptible to local lesion infection is resistant to systemic infection. Similarly, one resistant to local infection is susceptible to systemic infection. Sixty-eight bean varieties were tested and all were susceptible to either local or systemic infection. The varieties susceptible to local infection can be considered commercially resistant. The Sieva Lima bean and soy bean were the only other susceptible hosts found among 31 species representing 20 genera in 9 families. The virus was inactivated between 70° and 75° C. when heated for 10 minutes and at a dilution of 1–100,000. It resisted aging for 62 days at 18° C., but after 93 it was inactive.

Shiny pod (greasy pod) virus and its identity with black root virus. ZAUMEYER, W. J., AND H. REX THOMAS. A new virus of beans, formerly known as greasy pod and now called shiny pod virus, was described in 1947 as a strain of common bean mosaic virus. It was reported as infectious only to varieties susceptible to common bean mosaic. The identity of shiny pod virus and black root virus has now been established. Varieties such as Idaho Refugee, Logan, and Rival whose resistance to common bean mosaic is inherited from Corbett Refugee develop typical black root symptoms when inoculated with shiny pod virus. In contrast, varieties such as Great Northern 15 and 123 and Red Mexican 3 and 34 whose resistance to common bean mosaic is inherited from Great Northern 1 are resistant. Similarly, Robust does not develop either black root or shiny pod symptoms. Certain pole beans, such as Kentucky Wonder and Blue Lake, develop black root symptoms when inoculated with shiny pod virus. These studies show that black root symptoms develop only on certain varieties resistant to common bean mosaic, and shiny pod symptoms only on those susceptible to it. Although shiny pod virus is widespread in many western bean-growing areas on varieties susceptible to common bean mosaic, black root infection has been found only infrequently on varieties resistant to common bean mosaic, except those deriving their resistance from Great Northern 1.

Pythium seedling rot and root necrosis of Allium cepa. SEMENIUK, G., and B. N. WADLEY. Throughout a growing season *Pythium* spp. attack roots of onion plants grown from seeds, sets, or mature bulbs in peat or mineral soils in Iowa. Diseased roots plated on agar consistently yield high percentages of *Pythium* of spherical and nonspherical sporangical types, including *P. irregulare*, *P. ultimum*, *P. spinosum*, *P. mamillatum*, *P. debaryanum*, *P. graminicolum*. *Fusarium* develops from nearly every infected root, frequently with bacteria and nematodes; less frequent are a chlamydosporous *Phoma* sp., *P. terrestris*, and other fungi. Symptoms of Pythium parasitism are seed rotting, pre-emergence seedling rot, stunted emerged seedlings which often die, seedling damping-off, and lead-colored or faintly brown, yellowish or pink roots lacking cortex in advanced stages of disease. Nearly 100 per cent of pink roots from mineral and peat soils yield *Pythium* spp. with *Fusarium*, occasionally with *Phoma* and other fungi. Isolation techniques for *Phoma terrestris* eliminated *Pythium* spp. Pink root disease of onion in Iowa is primarily a Pythium root necrosis accompanied by *Fusarium* and *Phoma terrestris*.

FLUORINE ANALYSIS OF ITALIAN PRUNE FOLIAGE AFFECTED BY MARGINAL SCORCH

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INTRODUCTION

A new pathological condition, herein referred to as marginal leaf scorch, was reported in Italian prune trees in several localities in western Washington during the middle to latter part of the growing season following production of aluminum in those areas (Fig. 1). The injury was investigated in

Fig. 1. Part of Washington showing locations where samples of prune leaves were collected for fluorine analyses. Solid circles indicate location of aluminum factories.

1944, 1945, and 1946. It was found to occur in the areas of Washougal, Vancouver, Longview, and Tacoma in 1944 and 1945. In 1946 the leaf scorch occurred in the areas of Vancouver and Longview, but not near Washougal and Tacoma, following closure of the aluminum factories in the latter places. Fluorosis in cattle had been diagnosed in the area of each aluminum factory in Washington and the one at Troutdale, Oregon.

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All prune trees in an orchard usually within an area of 10 miles or less, but varying considerably with each factory in question, may be affected to some degree. Neither soil type nor cultivation greatly altered the condition, although the symptoms decreased with abundant water supply. All attempts to isolate an organism as a causative agent failed, and examination of indicator plants eliminated sulfur dioxide as a causative agent.

SYMPTOMS

The first symptoms on Italian prune trees appear as water-soaked areas around the leaf margin, or less frequently they may appear in the leaf lamina. The affected tissue darkens to a brown and may fall out (Fig. 2).

FIG. 2. Italian prune leaves showing different amounts of injury. Leaf at left shows marginal scorch only; two leaves at right have symptoms of marginal scorch and leaf spot. Samples collected near Washougal, Clark County, Washington.

All leaves may not be affected, but usually the lower half of the tree is more severely injured than the upper part. Where leaf scorch is severe as much as 50 per cent of the leaves, especially from the lower branches, may fall by the first part of August. In 1943 in the Washougal area the injured trees dropped most of the fruit two to three weeks before harvest and failed to set fruit in 1944 and 1945. A somewhat similar leaf condition in an apricot and fig orchard near an aluminum factory in California was reported to be caused by fluorine (8).

The production of leaf scorch in Italian prunes and other plants by fumigation with hydrogen fluoride in a sealed chamber has been reported by the

authors (16). A detailed report of this part of the work will be the subject of a subsequent paper.

LITERATURE REVIEW

Fluorine compounds may occur in the smoke from industrial plants using fluorides in their processes. Injury to vegetation by fluorine compounds has been reported in the neighborhood of aluminum, beryllium, copper, iron, ceramic, glass, brick, superphosphate, and miscellaneous chemical factories (3, 4, 5, 13, 14, 15, 23 through 27, 29, 30). The fluorine-containing compounds have been reported to be hydrogen fluoride (HF), hydrofluosilicic acid (H_2SiF_6), and silicon tetrafluoride (SiF_4) (5, 13, 14). These compounds, when present in the atmosphere even in very minute concentration, have the property of being absorbed by moisture droplets, such as dew or fog, to increase very greatly the concentration in a limited area (2, 14, 18, 26, 27, 29). In solution in dew hydrogen fluoride acts as a caustic acid. When the dew evaporates gaseous hydrogen fluoride may enter the leaf as a caustic gas (13). Thus the poisonous action of hydrogen fluoride, as both acid and gas, may be accomplished although the atmospheric concentration is very low. In a closed room injury to vegetation was observed following a three- to four-week exposure when the atmosphere contained as little as two p. p. m. of hydrogen fluoride (13). Under somewhat different conditions two p. p. m. of silicon tetrafluoride caused injury much more quickly (30), as did higher concentrations of either material (13, 30).

Descriptions have been given of the symptoms of injury from fluorine-containing gases for a number of species of trees, together with detailed microscopic study of changes in leaf structure (5, 13, 14). The description of the macroscopic injury to prune leaves is similar to that observed in western Washington. However, several investigators have pointed out that diagnosis of fluorine injury to plants cannot be proved by symptoms alone, since other factors such as climatic conditions and other industrial fumes may cause similar symptoms (5, 13, 14, 15, 17, 24). Chemical analysis of the injured tissue was used as the final proof of fluorine injury by these investigators.

The normal fluorine content of air-dried plant material, exclusive of roots, grown under normal conditions recently has been reported to be from 0.2 to 4.5 p. p. m.; fertilization of the soil with fertilizers containing fluorine has a negligible effect on the fluorine content of the foliage (6, 12, 13, 19, 20, 21, 22, 26, 28). However, a report from French Morocco (10) indicates that where ground-water fluorine is high, the fluorine content of some plants may be higher. Earlier reports (11) that dried plants normally contain 130 p. p. m. or more of fluorine are believed to be much too high, possibly due to inaccurate chemical methods available at that time for the determination of fluorine (5, 7).

Considerable variation has been reported in the susceptibility of different plants to fluorine compounds in the atmosphere. Hydrangea and tomato were reported more resistant than *Asparagus sprengeri* Regel., *Cyclamen sp.*,

and *Solanum capsicastrum* Link. (9). Ash has been reported to be more susceptible than several species of oak, maple, and beech (6); while conifers, such as pine and fir, are very easily injured (6, 14, 30). Very sensitive to hydrogen fluoride fumigation of 10 p. p. m. were willow, birch, sweet cherry, nut, plum or prune trees (Pflaumenbaum), potato and grape vines. More resistant were sour cherry and apple trees. The pear tree was very insensitive, and almost as resistant were locust, alder, oak, poplar, and elm (14). Under field conditions it was reported that beech, oak, linden, chestnut,

TABLE 1.—*Fluorine content of prune leaves collected in 1944, 1945, and 1946 from several areas in Washington where aluminum factories were in operation at time of sampling*

Location of sample (area)	Location of nearest aluminum factory	Distance from nearest aluminum factory		Sample date (approximate)	P. p. m. F in dry matter			Number of samples analyzed
		Miles	Direction		From	To	Av.	
Vancouver	Vancouver	0–1		[a]	137	199	168	2
		2–5	N to E	[b]	21	62	39	14
		5–10	N to E	[b]	13	80	30	11
		over 10	N to E	[b]	10	25	19	11
Portland (Ore.)[c]	Vancouver	3–6	S	[b]	88	132	110	2
Washougal	Troutdale (Ore.)	2	NE	Oct. 15	231	.....	.....	1
		2–5	N to NW	[b]	30	37	34	2
		2–5	NE to E	[b]	83	161	121	7
		5–10	NW.	Sept. 1	21	31	26	2
		over 10	NW	Sept. 1	18	.....	.....	1
Longview	Longview	0–3	W to NE	Oct. 15	61	65	63	2
		0–3	E to SE	[b]	110	420	274	13
Tacoma	Tacoma	0–3	N to SE	Nov. 1	130	1400	496	8
Whatcom and Skagit Counties	Tacoma	over 90	N	Oct. 15	6	15	9	5
Benton County	Troutdale (Ore.)	over 100	E	Oct. 1	8	.....	.....	1

[a] Samples taken approximately Oct. 15 and June 15.
[b] Samples taken at various times after July 15.
[c] Obtained from J. S. Wieman, Oregon State Department of Agriculture, Portland, Oregon.

walnut, grape, Damson plum, Myrobalan and Yellow Egg plum showed injury from fluorine-containing gases, while locust, maple, and blackberry showed only a trace of injury and apple and cherry were uninjured (14). The storage of fluorine in the young bark of trees exposed to fumes containing fluoride has been reported, but the translocation of fluorine into the new leaf growth did not occur (6). Thus scorch on new young leaves cannot be caused by stored fluorine (6).

MATERIAL AND METHODS

Leaf samples showing injury were collected from several localities in Clark County, Washington, where disease symptoms were acute, and from localities where the characteristic injury was slight or absent. In addition,

TABLE 2.—*The effect of operation of aluminum factories on fluorine content of Italian prune leaves*

Location of sample (area)	Location of nearest aluminum factory	Distance from nearest aluminum factory (miles)	Av. p. p. m. F in dry matter		Number of samples	
			1945	1946		
			Aluminum factory			
			Operating	Not operating	1945	1946
Washougal	Troutdale (Ore.)	2–5	121[a]	9[b]	7	5
Tacoma	Tacoma	0–3	496 (Nov. 1)	20 (Oct. 7)	8	2
			Operating	Operating		
Vancouver	Vancouver	2–5	35[a]	45[b]	8	6
Longview	Longview	0–3	303 (Oct.–Nov.)	179 (July 25)	10	3

[a] Samples taken at various times after July 15.
[b] Samples taken at various times after June 15.

prune leaf samples were also collected in the vicinities of Longview and Tacoma where leaf scorch was also prevalent. To determine the fluorine content of prune leaves in a disease-free area, leaf samples were collected in the northwestern and central parts of the State, ninety miles or more from the nearest aluminum factory. In all cases the samples were dried in an oven at about 75° C., ground in a Wiley mill, and analyzed for fluorine according to the method recommended by the Association of Official Agricultural Chemists (1, 7). Samples of twigs taken during the dormant season were divided into the preceding season's growth and earlier growth. They were dried, ground, and analyzed by the same procedure as that used for the leaves.

CHEMICAL ANALYSES

The results of the chemical analyses of leaves are shown in table 1. The data recorded show that in areas near the aluminum factories the fluorine content of the leaves was higher than in distant areas. This was especially

TABLE 3.—*Fluorine content of prune twigs of different ages*

Location of sample	Distance from nearest aluminum factory (miles)	Age of twigs and Av. p. p. m. F		Total variation	No. samples for each age
		1 year	2–3 years		
Clark County	2.5 –10	2.2	2.6	1.3–5.0	3
Tacoma	0.25	17.0	16.0	.....	1
	0.50	9.3	6.6	.....	1
	0.5 – 3.0	4.1	3.5	1.9–4.7	3
Whatcom County	100	0.7	1.0	0.5–1.1	2

true to the east and south of the factories but considerable variation occurred within an area.

The results in table 2 show the effect of the operation of aluminum factories on the fluorine content of Italian prune leaves. The fluorine content of prune leaves in the area of closed factories returned to approximately normal.

In order to determine if fluorine is stored in the tree, samples of twigs were taken in the dormant season and analyzed. The results of these analyses are shown in table 3. Although some fluorine was stored in the twigs and branches of injured trees, there was up to 100 times as much fluorine in the leaves.

DISCUSSION

In the Washougal and Vancouver areas where the summer rainfall is light, the severity of the injury was accentuated. There definite leaf scorch appeared with as little as 25–30 p. p. m. of fluorine in the leaves. It was observed in one orchard that trees watered by a sprinkler showed less injury than adjacent non-irrigated trees, although leaf samples from both trees contained 83 p. p. m. of fluorine.

In 1945 prune leaves from the area near Vancouver generally contained less fluorine than those in the vicinity of Camas and Washougal. The injury around the latter places fanned out in a wide area north of the factory at Troutdale, Oregon. That year the fluorine content of the leaves was higher northeast and east of the factory than was recorded at comparable distances to the northwest. The pattern of the injury around Vancouver was more general. All samples from the Longview area were taken close to the factory in question and were relatively high in fluorine and showed considerable marginal scorch. The same was true of the Tacoma area in 1945. In contrast, however, in 1946 when the factories were closed, the leaves did not show marginal scorch and contained 25 p. p. m. or less of fluorine. The same was true in the vicinity of Washougal. These results in general confirm the observation of previous investigators that fluorine is not translocated to new leaf tissue (6). In the areas around Vancouver and Longview, the amount of fluorine in the leaves and the degree of injury was similar in 1945 and 1946.

The samples taken from Whatcom, Skagit, and Benton Counties, which are over 90 miles from aluminum factories, may be considered to contain approximately the normal amount of fluorine for factory-free areas in Washington.

SUMMARY

A new non-parasitic disease of Italian prune trees characterized by marginal scorch and leaf spot appeared in certain areas of western Washington. In an investigation of the cause of the condition, it was shown by chemical analysis that leaves from trees grown in the affected areas were abnormally high in fluorine, as compared with leaf samples collected from other locali-

ties where this condition does not occur. The analyses from six samples collected 90 miles or more from an aluminum factory ranged in fluorine content from 6 to 15 p. p. m., while 76 samples from areas within 20 miles of an aluminum factory varied between 30 and 1400 p. p. m. of fluorine.

Within a given area the amount of marginal scorch was approximately proportional to the fluorine content of the leaves. The fluorine content of the prune leaves returned to near normal and marginal scorch of the leaves did not appear during the growing season following the closure of the two aluminum factories in Tacoma and Troutdale, Oregon.

The evidence indicates that some fluorine is stored in dormant twigs, but is not translocated to any great extent into the growing leaves.

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LITERATURE CITED

1. Changes in official and tentative methods of analysis made at the fifty-eighth annual meeting, October 27, 28, 1943. Section XXIX, Metals in Foods. Jour. Assoc. Official Agr. Chem. **27**: 90–100. 1944.
2. ALKE, A., and V. GRIMM. The fog catastrophe in the industrial territory south of Luttich. Münch. med. Woch. **160**: 283. 1931. Abstr. in Chem. Abstr. **26**: 1044. 1932.
3. BIRAGHI, A. [Determination of the presence of fluorine in vegetable tissue affected by gaseous fumes.] Boll. Staz. Patol. Veg. **18**: 360–366. 1938. Abstr. in Chem. Abstr. **33**: 9360. 1939.
4. BLAKEMORE, F. Industrial fluorosis of animals in England. Proc. Nutrition Soc. (Eng. & Scot.) **1**: 211–212, discussion 212–215, 1944. Abstr. in Chem. Abstr. **39**: 3836. 1945.
5. BREDEMANN, G. and H. RADELOFF. Zur Diagnose von Fluor-Rauchschäden. Phytopath. Zeit. **5**: 195–206. 1932.
6. ———— and ————. Über Fluor-Rauchschäden. Angew. Bot. **19**: 172–181. 1937.
7. CLIFFORD, P. A. Report on fluorine. Jour. Assoc. Official Agr. Chem. **28**: 277–279. 1945.
8. DE ONG, E. R. Injury to apricot leaves from fluorine deposit. Phytopath. **36**: 469–471. 1946.
9. ECKSTRAND, H. Toxicity to plants of a fluorine-containing wood preservative. Medd. Växtskyddsanst. Stockh. **36**: 1941. Abstr. in Rev. Appl. Mycol. **22**: 119. 1943.
10. GAUD, M., A. CHARNOT, and M. LANGLAIS. Le darmous humaine. Bull. Institut. Hyg., Maroc. Nos. 1 and 2. 1934.
11. GAUTIER, A., and P. CLAUSMANN. Action of fluorine upon vegetation. Field experiments. Compt. rend. **169**: 115–122. 1919. Abstr. in Chem. Abstr. **13**: 3267. 1919.
12. HART, E. B., P. H. PHILLIPS, and G. BOHSTEDT. Relation of soil fertilization with superphosphate including rock phosphate to the fluorine content of plants and drainage water. Amer. Jour. Pub. Health **24**: 936–940. 1934.
13. HASELHOFF, E. Grundzüge der Rauchschädenkunde. Gebrüder Borntraeger, Berlin. 1932.
14. ————, G. BREDEMANN, and W. HASELHOFF. Enstehung, Erkennung, und Beurteilung von Rauchschäden. Gebrüder Borntraeger, Berlin. 1932.
15. HÖRICHT, W. Waldverwüstung durch Rauch. Kranke Pflanze **15**: 90. 1938.
16. JOHNSON, F., V. L. MILLER, and D. F. ALLMENDINGER. Hydrogen fluoride injury to prune trees in Washington. (Abstr.) Phytopath. **37**: 362. 1947.
17. KRÜGER, E. Detection of fluoride in plants injured by fluorine. Metall. und Erz **38**: 265–266. 1941. Abstr. in Chem. Abstr. **36**: 4054. 1942.
18. MACHLE, W., F. THAMANN, K. KITZMILLER, and J. CHOLAK. The effects of the inhalation of hydrogen fluoride. I. The response following exposure to high concentrations. Jour. Ind. Hyg. **16**: 129–145. 1934.
19. MACINTIRE, W. H., S. H. WINTERBERG, J. G., THOMPSON, and B. W. HATCHER. Fluorine content of plants. Ind. and Eng. Chem. **34**: 1469–1479. 1942.

20. MCCLURE, F. J., and C. A. KINSER. Fluorine in domestic waters and its systemic effects. II. Fluorine content of urine in relation to fluorine in drinking water. U. S. Pub. Health Repts. **59**: 1575–1591. 1944.
21. MEYN, A. and K. VIEHL. Chronic fluorine poisoning in cattle. Arch. wiss. prakt. Tierheilk. **76**: 329–339. Abstr. in Chem. Abstr. **36**: 5259. 1942.
22. PHILLIPS, P. H., E. B. HART, and G. BOHSTEDT. Does fluorine in phosphorus fertilizers endanger the public health? Wis. Agr. Exp. Sta. Bull. 430: 125–129. 1935.
23. REYNOLDS, D. S., and R. D. JACOB. Volatilization of fluorine in manufacture of phosphorus and phosphoric acid by the furnace processes. Ind. and Eng. Chem. **25**: 1321–1323. 1933.
24. RECKENDORFER, P. Über den Nachweis von Fluor in Pflanzen und Bodenproben. Mikrochemie **3**: 126. 1931.
25. ROHOLM, K. Fog disaster in the Meuse Valley, 1930: a fluorine intoxication. Jour. Ind. Hyg. and Toxicol. **19**: 126–137. 1937.
26. ———. Fluorine intoxication. H. K. Lewis & Co. Ltd., London. 1937.
27. SCHWARZ, F. Maastel fog catastrophe and air-raid precautions. Protar **4**: 53–55. 1938. Abstr. in Chem. Abstr. **35**: 6013. 1941.
28. SMITH, H. V., M. C. SMITH, and M. VAVICH. Fluorine in milk, plant foods, and foods cooked in fluorine-containing water. Ariz. Agr. Exp. Sta. Mimeographed Rept. 77. 1945.
29. WILK, L. Injuries to crops and vegetation due to fumes from carbide and aluminium factories. Arch. Chem. Mikros. **9**: 176–189. 1916. Abstr. in Chem. Abstr. **15**: 3864. 1921.
30. WISLICENUS, H. Experimentelle Rauchschäden. Abstr. in Sammlung von Abhandlungen über Abgase und Rauchschäden. **10**: 32. 1914.

SOIL FUMIGATION FOR FUNGUS CONTROL WITH METHYL BROMIDE

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In the search for volatile fumigants that can be used to control not just nematodes but damping-off fungi, root and stem rots, and sclerotia of *Sclerotinia*, the writers' experience with methyl bromide seems worth recording at this time. The results of five tests indicate that the ease with which methyl bromide can be handled, the absence of lachrymating and of dangerous phytocidal properties and its rapid penetrating power recommend it to some florists and vegetable plant growers who want a quick, labor-saving way to treat their seed bed or potting soil and are willing to pay a premium for a less disagreeable and quicker method than formaldehyde, steam, or chloropicrin can offer.

Methyl bromide has been employed in several ways in different experiments. The pure liquid, which comes in pound cans and cylinders under pressure at room temperatures, has been liberated in soil placed in 55-gallon, covered, metal drums. In a similar manner one- and two-cubic-yard piles of soil have been treated under a gas-proof tarpaulin. Solutions of methyl bromide on the market as Dowfume G and Iscobrome have also been used by injection methods at one-foot intervals in low piles or lesser intervals in flats of soil and immediately sealed in. One grower has been applying one of the above products by a sprinkler technique, at intervals over the soil, while building his compost pile with a steam shovel.

In 1945 comparative tests made by the junior author with several fumigants in gallon crocks of soil infested with species of *Pythium* and *Fusarium* showed that as the dosage rates of Dowfume G and Iscobrome were increased well above those needed for nematode control, the control of damping-off of peas was improved. With this and other clues as to the possible efficacy of methyl bromide as a soil fungicide, two 55-gallon metal drums were filled with a compost soil obtained from a florist in Rochester. The soil in one had a temperature of 48° F. and in the other 60° F. at the time of treatment. The drums had rubber gaskets and tight-fitting lids. Soil moisture was adequate for good seed germination. Exactly 90 ml. of methyl bromide was metered into the center of each drumful of soil through "Saran" tubing inserted through a rubber stopper.[1] Since there were 8 cu. ft. of soil in each drum, the amount of methyl bromide used was approximately 11 ml. per cu. ft. The drums were held in rooms at approximately 48° F. and 60° F., respectively, for 6 days after which samples of soil both treated and untreated were placed in flats and sown with spinach and beet seeds. The results are given in table 1. The grower used the balance of

[1] A convenient metering device for methyl bromide is made by the Arrow Products Company, Carlstadt, N. J.

TABLE 1.—*Results of soil treatments in metal drums*

Treatment	Average Percentage germination		Number of weeds
	Spinach	Beets	
Check	2	16	27
Methyl bromide 90 cc.—48° F.	62	88	4
Methyl bromide 90 cc.—60° F.	45	93	0
Steamed 30 min. at 15 lb. pressure	50	100	0

the soil to raise snapdragon seedlings. These are very subject to damping-off but for the first time in years he had practically no loss and almost no weeding to do.

In August, 1945 Stark and Lear treated two compost piles comparing a 10 ml. per cubic ft. rate of chloropicrin (Larvacide) with a 50 ml. rate of Dowfume G. The soil temperature was 75° F. Injections were made 3 inches deep and the piles sprinkled with water after which the chloropicrin-treated pile was covered with wet canvas and the Dowfume-treated one

TABLE 2.—*Results of compost pile treatment with Larvacide and Dowfume G*

Treatment	No. of weeds	Stands	
		Cucumbers Per cent	Spinach Per cent
Check	1287	54	16
Larvacide at 10 ml./cu. ft. Pile sprinkled and covered with wet canvas	14	87	64
Dowfume G at 50 ml./cu. ft. Sprinkled and covered with gas-proof tarpaulin	3	80	76

with a gas-proof tarpaulin. At the end of 5 days, samples of treated and untreated soil were sown to spinach and cucumbers. The final stands of seedlings and of weeds are given in table 2 from which it can be seen that under these conditions, methyl bromide performed as well as chloropicrin both as fungicide and herbicide.

The importance of adequate confinement of the fumigant was indicated by a similar experiment in September where liquid methyl bromide was

TABLE 3.—*Results of compost pile treatments with methyl bromide*

Methyl bromide treatment	No. of weeds per flat	Average percentage stands (3 flats)		
		Spinach	Cucumber	Sweet pea
Check—None	114	20	25	20
½ lb. per cu. yd. (under gas-proof tarpaulin)	0	18	51	29
1 lb. per cu. yd. (under gas-proof tarpaulin)	0	43	80	60
1 lb. per cu. yd. (under canvas cover, not gas-proof)	21	10	20	27

discharged from a one pound can under different coverings into 1 and 2 cu. yd. piles of compost soil at Auburn, N. Y. Soil temperature was only 56° F. The results, as measured by seedling growth in samples of the soil, are given in table 3, and are shown in figure 1.

Again 12 ml. methyl bromide per cu. ft. was adequate at the rather low temperature of 56° F. There is reason to believe from the work of the U. S. Bureau of Entomology that at soil temperatures about 20° F. higher a 25 to 50 per cent reduction of this dosage might be adequate under proper conditions of confinement.

On February 11, 1947, a quantity of sandy loam compost heavily infested with weed seeds and damping-off fungi was brought indoors from cold frames and treated in a layer 1 foot thick on a concrete floor in a farm building near Rochester, N. Y. The soil moisture was fairly high but temperature was only 52° F. Injections of 60 ml. of Dowfume G were made on 12-inch centers about 5 inches deep. A gas-proof tarpaulin was

TABLE 4.—*Compost pile injections with Dowfume G*

Crop sown	Mean percentage stand of four replicates		Total no. weeds (2 flats)	
	Untreated	Treated	Untreated	Treated
Beets	60	95	225	72
Cucumber	46	83	237	104
Peas	7	94	462	176[a]
Tomatoes	42	66		
Spinach	10	32		
	165	370		

[a] All weeds in treated flats turned out to be clover while purslane and grasses were also present in checks.

used as cover and left on 48 hours. Samples of treated and untreated soil were brought to Ithaca and sown with 5 different vegetables. Results of this test are given in table 4, and may be seen in figure 2.

The grower made a similar test of treated and untreated soil using lettuce as an indicator, results of which were equally striking (Fig. 2, B), namely a 50 per cent increase in stand with a corresponding decrease in weeds due to treatment. The same grower, the year before, had treated part of his soil in the metal drums using Iscobrome, and noted much better growth of his melon seedlings.

In another test,[2] in which Dowfume G was compared with steam, formaldehyde, and chloropicrin, soil was treated *in situ* in flats ($14 \times 18 \times 3$ inches) with the methyl bromide fumigant by pouring 5 ml. in each of 4 spaced holes ($1\frac{1}{2}$ inches deep). The treated soil was immediately sealed by sprinkling lightly with water, then the flats were covered with glue-coated paper and stacked in piles for two days at approximately 75° F. In other stacks were steamed, chloropicrin-treated, and formaldehyde-treated flats. The stands of seedlings are given in table 5.

[2] Test made by students in plant disease control course taught by Dr. L. J. Tyler.

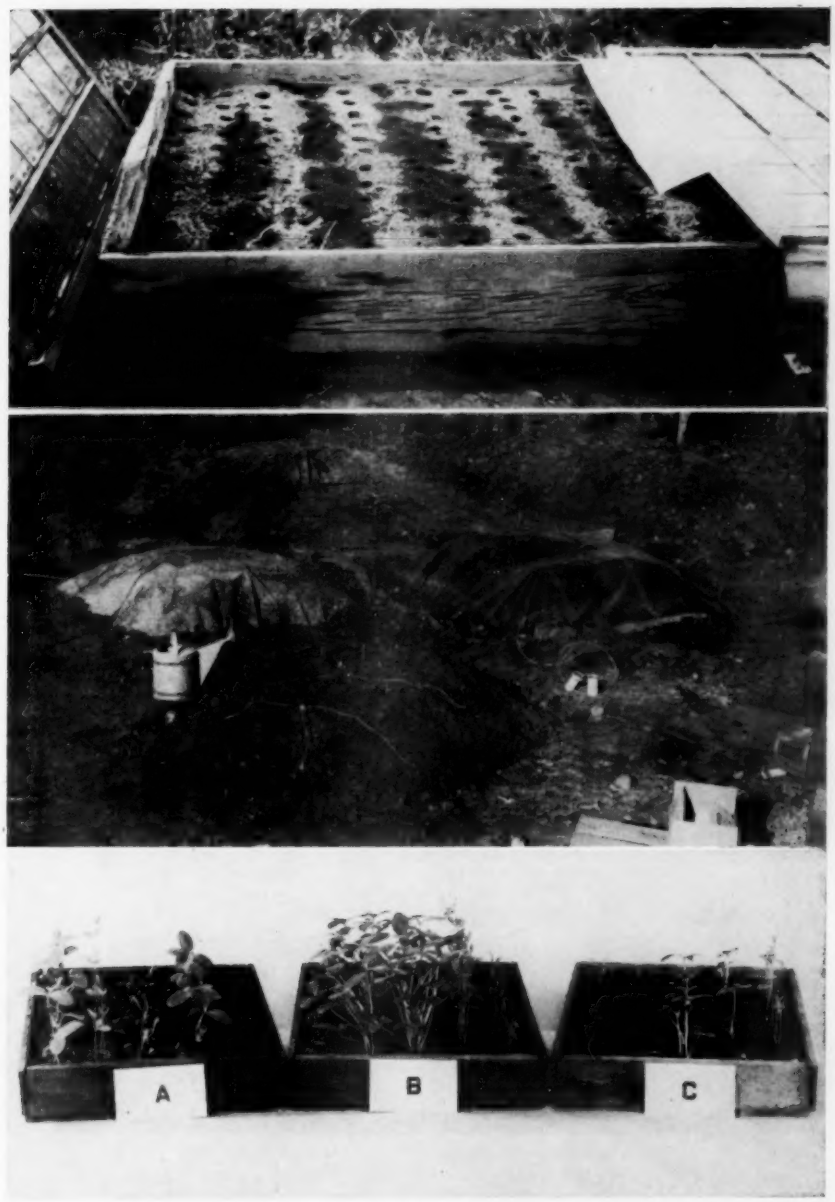

FIG. 1. *Top:* Coldframe treatment with straight methyl bromide showing holes punched in soil at 6- to 8-inch intervals, method of covering with glue-coated paper, and three strings attached to buried sclerotia. *Center:* One- and two-cubic-yard piles of compost soil being treated with one-pound cans of methyl bromide under different types of cover. Strings are attached to bags of buried sclerotia. *Bottom:* Some results of treatments given above. A, Check. B, one pound methyl bromide per cubic yard under gas-proof tarpaulin. C, same under ordinary water-proof tarpaulin. (Mere water seal was also inadequate.)

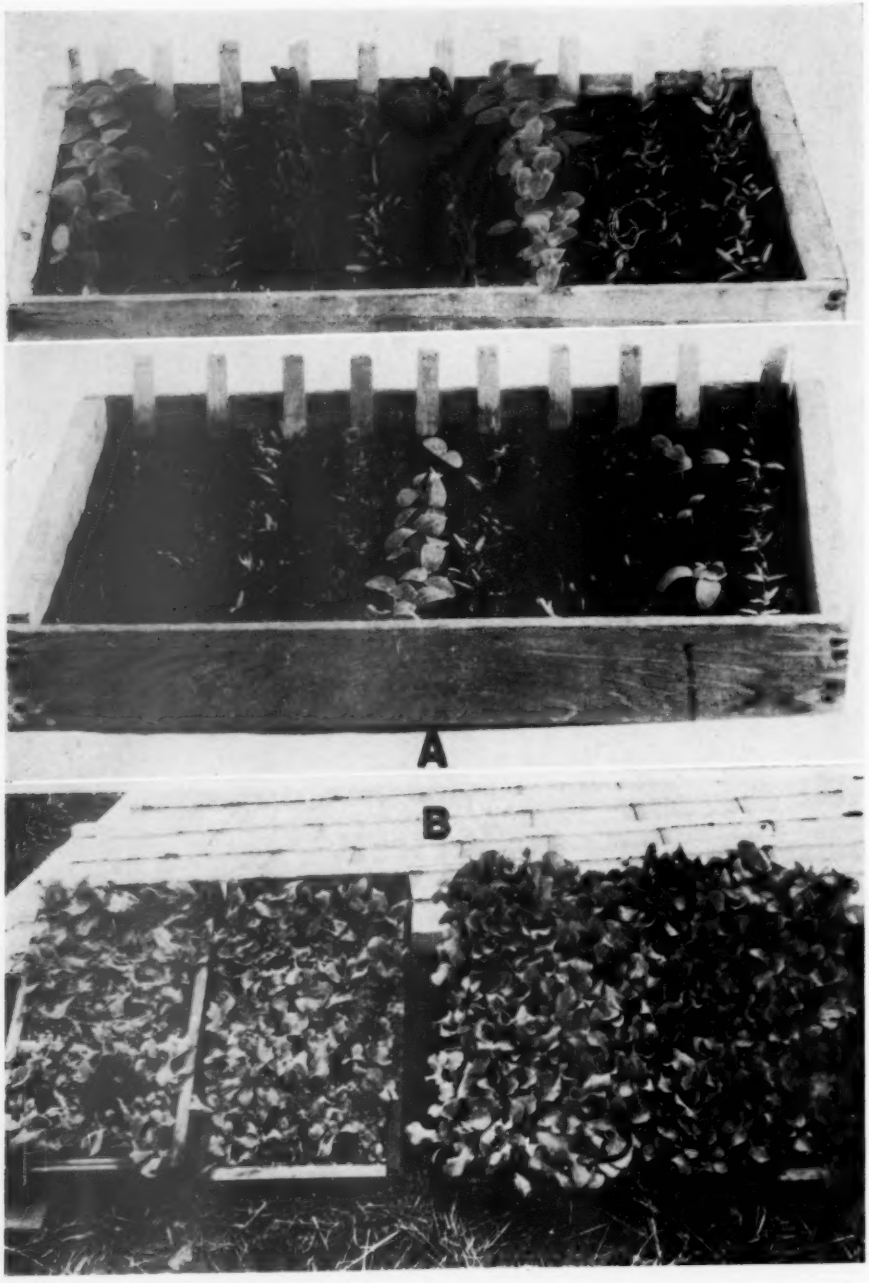

FIG. 2. Results of cold-frame soil treatment with Dowfume G at 60 cu. cm. injections on 12-inch centers covered 2 days with gas-proof tarpaulin. A. Upper flat treated, lower check. B. Two flats on left untreated soil, right treated.

At the rate employed, a pound of Dowfume G at 35¢ would treat 16 flats, at a cost for material slightly over 2¢ a flat. It is probable that several layers of wet newspaper could be successfully substituted for glue-coated paper as a wrap for the flats.

On three or four occasions the sclerotia of *Sclerotinia sclerotiorum* have been buried in soil which was being treated with either straight methyl bromide at 11 ml. per cu. ft. or with Dowfume G at 50 ml. per cu. ft. Even at soil temperatures between 50° and 55° F., complete kill has been obtained provided the sclerotia were soaked a few hours prior to fumigation. When they were dry, many of them survived the treatment regardless of soil temperature.

These methods of treating soils with either straight methyl bromide or with commercial preparations containing this fumigant will be found useful in situations where a grower wishes to obtain fungus control and is willing to pay a small premium in order to avoid the disagreeable, or the

TABLE 5.—*Results of treating flats of soil*

Crop sown	Final stands of seedlings (2 flats)				
	Check	Steam	Chloropicrin	Formaldehyde	Dowfume G
Cucumbers	5	28	25	33	32
Spinach	5	29	49	36	35
Tomatoes	16	17	19	23	31
Beets	25	115	98	117	106
	51	189	191	209	204

phytocidal drawbacks of chloropicrin fumigation or the labor and time required in the formaldehyde treatment of small quantities of soil. One advantage of using straight methyl bromide in cases where the soil can be held in a gas-proof drum or other container is the short time required for the entire operation. It is believed that a 12-hour treatment followed by a similar period of aeration will be found ample time to keep soil out of use, wherever seeds are to be sown. Further experiments may show that results can be obtained with lower dosage rates than the ones recorded here.

While methyl bromide so far has been tested against only damping-off fungi and the sclerotia of *Sclerotinia sclerotiorum*, it seems entirely possible the methods outlined may be effective in eradicating other pathogens. The potency of this fumigant against nematodes, when used in metal drums was pointed out by Taylor (1941)[3] who found 1 ml. per cu. yd. adequate. Since methyl bromide fumes are practically odorless and dangerous to man, the operator should always provide plently of ventilation when working around or with it.

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[3] Taylor, A. L. Chemical control of root-knot. Larvacide Log. 4: 63–66. Aug. 1941. Publ. by Innis Speiden Co., N. Y.

CLITOCYBE ROOT ROT OF CITRUS TREES IN FLORIDA

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(Accepted for publication September 19, 1947)

INTRODUCTION

Numerous cases of mushroom root rot, caused by *Clitocybe tabescens* (Scop. ex Fr.) Bres., have been recorded by the writer since 1924 on a large variety of woody plants from various parts of Florida, where the disease is known to have occurred as early as 1902 (14). In addition to attacking native forest trees, this fungus attacks tungoil trees and a large number of fruit trees and ornamental trees, shrubs and vines, including many exotic ones. It was not until the end of 1929, however, that definite evidence was secured that citrus trees also are attacked. At that time, specimens of the roots of rough lemon (*Citrus limon* (L.) Burm. f.) under grapefruit at Lake Alfred, Polk County, sent to the writer for diagnosis, were found attacked by root rot, and pure cultures of *C. tabescens* were obtained from them. An inspection of this grove early in 1930 revealed that this disease was extensive in the root systems of a considerable number of 15-year-old grapefruit trees. During this and succeeding years root rot was found in other citrus groves at Lake Alfred and in groves at a number of other points in Polk County, as well as in other counties in Florida. Preliminary reports of its occurrence have been presented by the writer (7 to 21).

Although the occurrence of Clitocybe root rot of citrus trees in Florida was not recognized until late in 1929, this disease undoubtedly has caused considerable loss of trees in groves for many years. One grower informed the writer that this was the disease for which he began treating in 1915 in his grove at Kathleen, Polk County, where it was still attacking trees in 1932. A further record of what clearly was this disease is afforded by notes, dated August, 1916, found several years ago in the files of the Florida Agricultural Experiment Station. These were based on a letter and specimens of the butt of a rough lemon under grapefruit sent by Mr. S. F. Poole of Lake Alfred. The writer has found Clitocybe root rot prevalent and destructive in a number of the groves at Lake Alfred formerly in Mr. Poole's charge. It is believed that both records constitute definite evidence that this disease occurred in the citrus groves of Polk County at least 15 years before its presence was demonstrated.

DISTRIBUTION AND ECONOMIC IMPORTANCE

No opportunity has been afforded to make a systematic survey of the citrus region of Florida to determine the exact distribution of Clitocybe root rot, though a number of groves at various points in Polk and Brevard

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Counties, where this disease was reported or suspected to occur, have been examined in more or less detail for 10 years, beginning in 1930. Observations in other counties have been largely incidental to other projects.

Unfortunately, even in groves where this disease occurs, unless fruiting bodies of the causal fungus develop, it is difficult to locate attacked trees without digging to expose the root systems for examination. In the absence of the fruiting bodies, attacked trees rarely have symptoms of the disease until the root systems have been killed to such an extent that the trees begin to decline, and such symptoms may be indicative of a number of troubles.

Clitocybe root rot of citrus trees has been found thus far principally in various sections of Polk County, where plantings are almost entirely on rough lemon stock. This disease also has been found to a limited extent in a few groves in Brevard County, where this rootstock is used but rarely. It also has been found occasionally in Lake, Marion, and Highlands Counties.

Clitocybe root rot has been found more or less frequently in Polk County in several groves in the vicinity of Lake Alfred, in a few around Winter Haven, in 3 in the Lakeland Highlands section, 2 near Waverly, 2 at Lake Hamilton, 1 at Kathleen, 1 near the south end of Lake Juliana 7 miles north of Auburndale, and 1 at Lake Wales. The prevalence of this disease at several more or less widely scattered localities in this county indicates that a more extensive survey will establish its occurrence at other points.

In Polk County, where Clitocybe root rot of citrus trees is most frequent and widespread, the incidence of the disease, where found, varied within wide limits. The first examination usually did not suffice to locate all the attacked trees in most of these groves, and in some from one to several additional ones were found at subsequent inspections. In some cases the attacked trees occurred in groups but more often they were scattered irregularly throughout the groves or parts of them, indicating various centers of infection.

At Lake Alfred the number of trees attacked varied from one or a few in 5 groves to at least 15 or 20 in another, and 40 in still another. A survey of 6 of these groves under one management,[2] supplemented by subsequent inspections in some, resulted in locating a total of 80 attacked trees, comprising 0.2 per cent of approximately 40,000 trees on about 700 acres. Since these groves average about 60 trees per acre, the total number of attacked trees would equal about 1⅓ acres out of the 700. These groves were from about 12 to 19 years old.

In the groves examined in the vicinity of Winter Haven the incidence of the disease mostly ranged from but a single tree to a few scattered ones, though in one grove visited later 30 trees about 15 years old were being

[2] The writer is indebted to Mr. S. F. Poole, who, prior to his death in 1936, was vice-president of the Florida State Horticultural Society and prominent grove owner and developer at Lake Alfred, for his valued cooperation and interest manifested in the location and treatment of trees attacked by root rot in the groves under his charge.

given surgical treatment after the disease had progressed so far that they had commenced to decline.

In the first grove inspected for root rot in 1931 at Highland City in the Lakeland Highlands section, where a considerable number of trees were reported to be dying, it was found that 34 out of a total of 384 on the first 16 rows inspected were attacked and had been given surgical treatment. Forty-eight trees in this 10-acre block had been removed on account of decline from root rot. An examination of the diseased roots from the 34 trees treated a short time previously showed that the trouble was due to Clitocybe root rot. In another 10-acre block, 14 years old, later examined in this section the owner stated that 8 per cent of the trees were attacked by this disease and that 28 trees had been given surgical treatment a short time prior to the inspection. This disease was found to occur frequently in another grove in this section that was examined by superficial digging around suspected trees, and growers have reported its occurrence subsequently in others.

Clitocybe root rot also occurred extensively in portions of a grove at Kathleen, and the former owner[3] reported that losses began about 1915, when the trees were about 5 years old.

A large grapefruit and an orange, both on rough lemon stock, were found attacked by Clitocybe root in a block of grove at the south end of Lake Juliana 7 miles north of Auburndale, in which several old seedling sweet orange trees having a number of dead and rotted roots had been treated surgically several years before for a disease thought at the time to have been foot rot (*Phytophthora parasitica*). Further evidence of the activity of Clitocybe root rot in this property was afforded by finding it causing the death of several trees in a commercial planting of guavas (*Psidium guajava*).

In Brevard County, the only other county in which Clitocybe root rot was found occurring to any particular extent, this disease was limited to a few groves in the central portion. It was most frequent in portions of two groves on rough lemon stock at Georgiana on Merritt Island. In one of these, where the trees were 15 years old, 38 trees in one corner were attacked. In the other, where the trees were 10 years old, 10 scattered trees were attacked. Instances of the disease on single trees on the same rootstock were found at Courtenay on Merritt Island, City Point, and Melbourne. The one at Melbourne was a calamondin in the yard of a residential property where a number of ornamentals also had died from the disease. This

[3] The writer is indebted to Mr. W. E. Tucker of El Paso, Texas, the former owner and developer of this grove, for the information so kindly furnished by him with respect to the occurrence of Clitocybe root rot and his experience with this disease in this grove. Correspondence with this grower began following the publication of an article by the writer (9), in which he reported the occurrence of this as a new disease of citrus trees in Florida. In May, 1932, Mr. Tucker wrote that he was convinced that this was the same disease that he had encountered and treated in his grove. He gave a diagram showing the varieties and rootstocks in the different blocks and the results of his experience with this disease. He stated that he had found it quite controllable, provided it was discovered and treatment administered before the root systems became seriously damaged. Further details of the occurrence of root rot in this grove are presented in the section of this paper dealing with rootstocks.

disease also was found to have attacked extensively the root systems of four seedling sweet orange (*Citrus sinensis* (L.) Osb.) trees about 45 years old in one part of a grove at Rockledge.

Records of the occurrence of Clitocybe root rot in Lake County consist of a seedling grapefruit (*Citrus paradisi* Macf.) tree probably 45 years old and a seedling sweet orange tree in a grove at Umatilla where trees had been lost over a period of years from a root trouble attributed to foot rot, and also a seedling sweet orange tree in an abandoned grove near the south shore of Lake Apopka, west of Oakland.

The only record of Clitocybe root rot in Marion County consists of a single seedling sweet orange tree in a grove in Orange Hammock west of Altoona, although the owner previously had reported losing an occasional tree from this trouble.

In the brief opportunity to examine citrus groves in Highlands County, the writer found only a single case of Clitocybe root rot on rough lemon under Temple orange at Lake Placid. However, judging by the prevalence of oak scrub prior to clearing and the death of large numbers of horsetail beefwood (*Casuarina equisetifolia*) trees from this disease in roadside planting, it should occur frequently in the citrus groves of this section.

A systematic survey will probably show Clitocybe root rot to be of fairly widespread occurrence throughout much of the ridge section of Florida, where rough lemon stock is used generally, and probably also in sections other than those here reported. This disease was reported by the county agricultural agent to occur in groves on sweet orange stock at De Land, Valusia County, but the writer has seen no cases in the few groves he has examined.

A total of approximately 340 bearing citrus trees, nearly all on rough lemon stock, have been found attacked by Clitocybe root rot in the preliminary observations thus far made in Florida.

SYMPTOMS

The symptoms in citrus trees attacked by Clitocybe root rot vary greatly according to the progress of the disease. As a rule symptoms of decline do not develop until the disease has been working for a number of years and has killed a large proportion of the root system. A surprisingly large number of attacked trees in groves in various localities of the State have been located merely by the presence of either fresh or old, dried clusters of the fruiting bodies of *C. tabescens* at the trees bases (Fig. 1) in cases where the disease had been developing for some time but had not progressed sufficiently to cause other symptoms. Such trees usually appear as healthy and productive as adjacent nonattacked trees, though sometimes showing a general lack of vigor and a slight yellowing of the foliage. However, upon removing the soil from the root crowns and adjacent roots, a number of the lateral roots are found invaded and killed for varying distances from the root crown (Fig. 2). In most of the attacked trees examined the taproots

also were invaded and killed. The disease generally appears to start at some point on the lateral roots or adjacent to the root crown and to spread

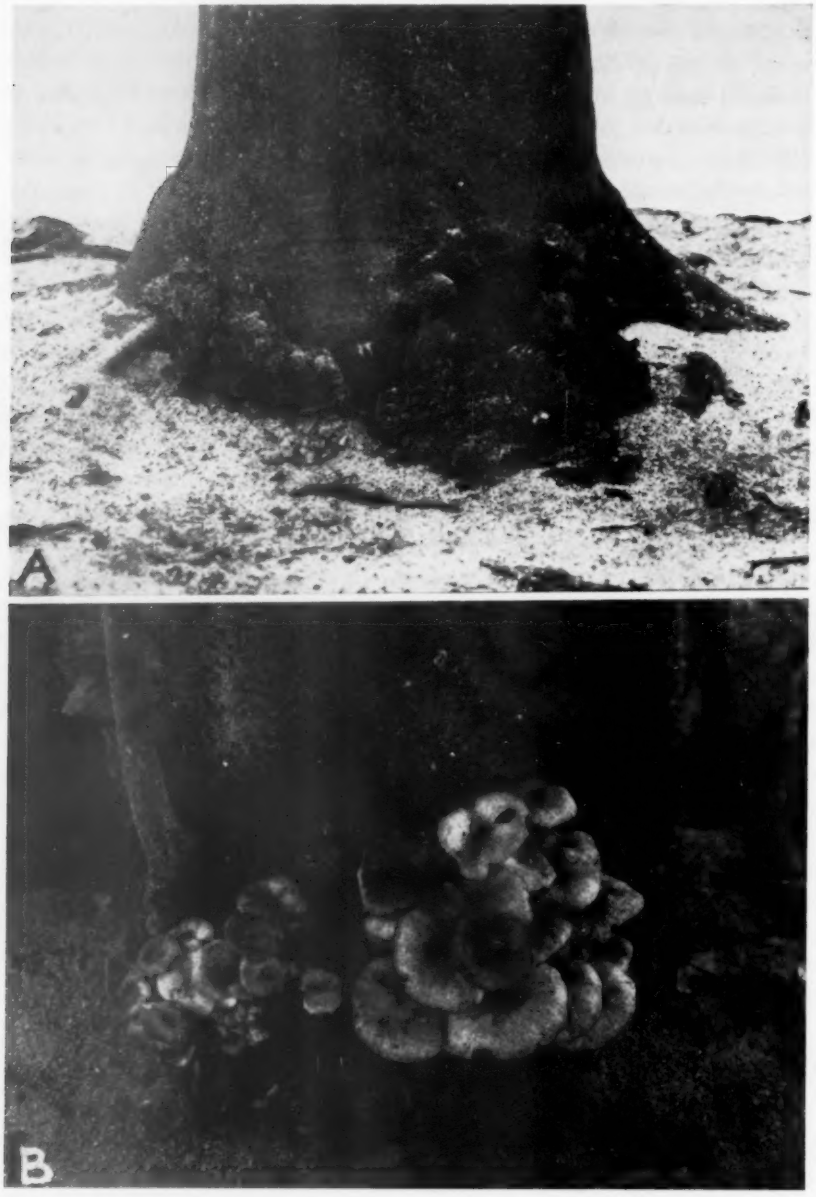

FIG. 1. Clusters of mushrooms of *Clitocybe tabescens*. A. Old clusters at base of grapefruit tree on rough lemon stock. B. Fresh clusters at base of orange tree on rough lemon stock.

thence to other roots and the base of the tree. The distal portions of attacked roots frequently are not invaded by the fungus and remain alive

until the roots are girdled. In some cases elongated bark lesions varying from a few inches to a foot or more in length have been found along one side of living lateral roots. In a number of cases such lesions had become delimited by the formation of a callus around the periphery, in which case the dead bark eventually disintegrated and sloughed off. In some cases a small dead area was found at one side of the base of the trunk. Such places frequently mark the point where a large lateral root has been killed back to the root crown and usually are places at which one or more clusters of the fruiting bodies of the fungus have developed.

In more advanced cases of the disease where fruiting bodies of the fungus were not present a further symptom sometimes is a slight gumming around

FIG. 2. Valencia orange tree on rough lemon stock attacked by Clitocybe root rot, showing young cluster of mushrooms developing at tree base and dead bark on root crown and roots after soil was removed.

the root crown and extending upward on the base of the trunk. The gumming area of bark dies within a short time and develops small longitudinal cracks. In still older cases the bark at the base may die on one side of the trunk, or sometimes partly around it, and crack loose along the callus developed at the margin where the bark remained living. These basal lesions usually extend upward from a few inches to 12 or 15 inches, and closely resemble lesions due to foot rot. In long-standing cases of root rot the bark of these lesions may be completely sloughed off. Not infrequently, these basal lesions serve as points for the entrance or fruiting of secondary wood-rotting fungi, particularly species of *Ganoderma*.

In the majority of the attacked roots of numerous citrus trees that have been examined, the mycelium of the Clitocybe root-rot fungus was abundant,

except in those cases where it had run its course and died. The mycelium varies from thin, white to cream- or chamois-colored wefts to felty layers of extensive development between the bark and the wood and also through the inner layers of the bark. Such mycelial sheets often are characterized by a more or less radiating or fan-shaped marginal development and a distinctly perforate effect. Where the margin of the mycelium is invading living roots a slight gumming may occur in the cambial region (Fig. 3). Freshly dug roots in which the mycelium of this fungus is still active have a pronounced mushroom or fungus odor.

In citrus trees the mycelium of the Clitocybe root-rot fungus rarely extends much above the ground-line, although it usually develops upward for distances ranging from a few inches to a foot or two in other attacked trees. Mycelium of this fungus has been found occasionally, however, between the

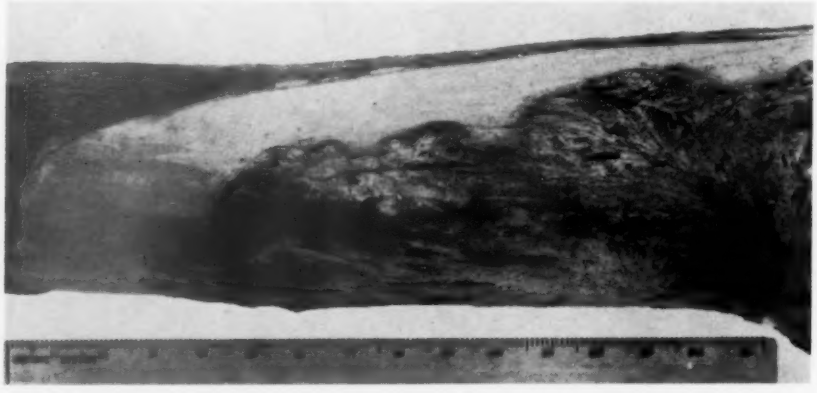

FIG. 3. Living root of old seedling sweet orange tree attacked by Clitocybe root rot, with bark cut away, showing radiate development of invading mycelial sheet and gum formation in cambial region.

bark and the wood in actively growing localized lesions at one side of attacked citrus trees. As a rule, though, it is rarely possible to diagnose this disease on citrus trees by the simple expedient of making a cutting test of the bark at the base to reveal the presence of mycelial sheets, as usually can be relied upon in other trees if they have been attacked long enough for the mycelium to extend upward between the bark and the wood above the ground-line.

Both radial and concentric cracks often appear in the wood of the larger attacked roots and become filled with whitish, felt-like layers of mycelium. In several instances even the wood in the butt of the tree has been invaded and heart rot has resulted, so that occasionally trunk divisions of large attacked trees split apart. In well-advanced cases of the disease the fungus causes a soft, whitish, delignifying type of decay of the deeper-lying roots and the decayed wood usually becomes watersoaked, soft and spongy, and often distinctly gelatinous in the late stages of decay. Longitudinal sections of such extensively decayed roots frequently show extensive pockets,

with whitish strands of mycelium in the interior, honeycombing the wood (Fig. 4).

The black, rounded or flattened, cortical and hypogeal, string-like rhizomorphs so frequently accompanying the closely related Armillaria root rot have never been found associated with root rot caused by *Clitocybe tabescens*, either in citrus or other trees. In both citrus and other trees attacked by *Clitocybe*, however, peculiar blackish, indurated xylostroma outgrowths, resembling the stromata of certain Pyrenomycetes, frequently develop through longitudinal fissures in the bark of attacked roots (Fig. 5). These extrusions do not develop in all cases but develop profusely in some.

FIG. 4. Longitudinal section through division of taproot of seedling sweet orange tree attacked by Clitocybe root rot, showing soft, spongy, delignifying type of decay and honeycombing of wood.

They may develop on both small and large roots but are much larger and more prominent on large roots, where they may be $\frac{3}{16}$ inch wide and several inches long. Such structures also occur more or less commonly in the root rot caused by *Armillaria mellea*.

In the case of a single seedling grapefruit and a few seedling sweet orange trees attacked by Clitocybe root rot, on roots with abundant xylostromata there also was noted black, bristle-like rhizomorphic structures protruding a short distance beyond the bark at right angles to the growth axis of the roots. These structures have been noted in but very few cases. Judging by their fineness, they would be very fragile when the soil becomes dry and would also be rather short-lived. The remains of these structures are apparent as black specks studding the bark between the network of

xylostroma extrusions (Fig. 5). In the case of a single old seedling grapefruit tree attacked by Clitocybe root rot at Umatilla, on a section of one root that was removed, a few scattered, flabby, black rhizomorphic structures were observed on the under side, extending downward into the soil. These had a maximum length of approximately half an inch.

By the time the Clitocybe root-rot fungus has invaded a portion of the root crown and sufficient of the roots to interfere with the maintenance of the top of the tree, the tree declines rapidly. The foliage becomes pallid and somewhat smaller than normal and the leaves become chlorotic along the midribs, as is invariably the case with acute starvation of citrus trees by any form of girdling or root-killing. The most seriously affected branches die back rapidly and the fruit produced on these devitalized branches is small and frequently drops before reaching maturity because of

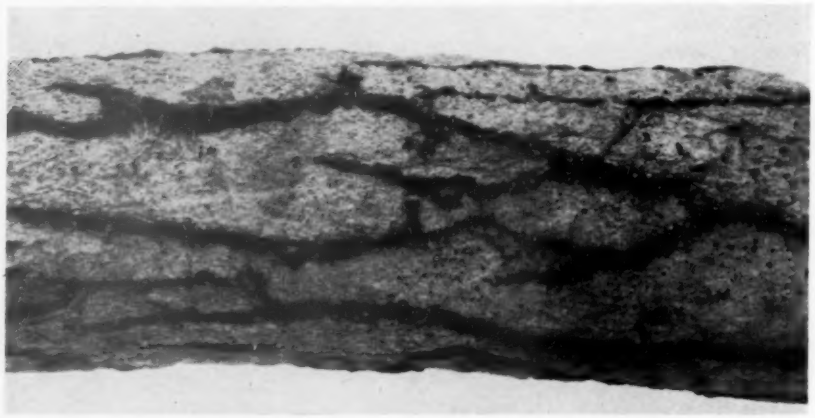

FIG. 5. Root of old seedling grapefruit tree attacked by Clitocybe root rot, showing blackish xylostroma outgrowths developed through fissures in the bark and the remnants of small, bristle-like, rhizomorphic structures protruding through the bark at right angles to it. Natural size.

the decline of the top. In the spring such declining trees develop an abnormally heavy bloom, which is also a characteristic feature of girdled or root-injured trees. In some cases where the bases of attacked trees become extensively girdled by the fungus the entire top may wilt and die rather rapidly. In some instances trees that have been greatly weakened by the rotting of a number of roots may be blown over by high winds.

CAUSE

The root-rot disease under discussion is caused by *Clitocybe tabescens* (Scop. ex Fr.) Bres., one of the gill fungi. The fruiting bodies of this fungus consist of a few to many individuals with the stems developing from a common base. When fully developed, the caps are convex to flattened or centrally depressed with age, whitish to light tan- or honey-colored, smooth or adorned with tufts of fibrils near the center, and from 2 to 3½ inches in diameter, with whitish gills underneath. The united *Clitocybe*, as this

fungus commonly is called, resembles the closely related honey agaric (*Armillaria mellea*) in habit of growth, color, texture, and general appearance but is distinguished principally by the complete lack of an annulus, dissimilar spores, and in being more slender from the beginning. *Armillaria* is of cosmopolitan occurrence and causes a very similar root rot. A comparative cultural study of these two closely related root-rot fungi has been made (19). *A. mellea* occurs more or less frequently in parts of northern Florida but in other parts of this State it is largely supplanted by *C. tabescens*. The mushrooms of the latter fungus usually attain their full development within a few days from the time the young "buttons" appear and decay rapidly in warm, rainy weather. However, if dry weather follows their development the clusters may dry and turn dark brown to blackish but remain recognizable for several weeks.

The presence of either fresh or old clusters of the fruiting bodies of the fungus frequently enables one to locate attacked citrus trees long before the presence of the disease is even suspected. In groves where Clitocybe root rot is known to occur a periodic inspection for the fruiting bodies of the causal fungus often will enable the grower to locate attacked trees and treat them before the disease kills the roots to such an extent that it becomes difficult or impossible to save the trees.

The production of fruiting bodies of the fungus depends largely upon the progress of the disease and the seasonal conditions. They develop with greatest frequency during the fall and early winter, from about mid-September to mid-December, but may develop in other months as well if soil moisture is favorable. As a rule only one or two clusters develop during the year and frequently none develop.

Origin and Anatomy of Xylostroma Outgrowths in Bark of Roots

The structure of the xylostromata that frequently develop as hard, blackish, stromalike structures extruded through more or less longitudinal cracks in the bark of roots of woody plants attacked by *Clitocybe tabescens* is essentially identical with that of the outgrowths of *Armillaria mellea* described and illustrated by various writers. The structure of the mycelial sheets of *Armillaria* as they occur under the bark was described by Hartig (5) as similar to that of the rhizomorphs except for modifications due to their different form and environment. Their occurrence in connection with the "collar crack" disease of cacao (*Theobroma cacao*) caused by *Armillaria* was described and illustrated by Dade (2). In the Gold Coast form of this disease on cacao in Africa, however, he found mostly a meager development of xylostromata between the bark and the wood but a strong tendency for development of xylostromata in the prominent medullary rays of the wood, which resulted in radial splitting of the stems. He designated the more or less frilly outgrowths of these mycelial sheets through longitudinal fissures in the bark as xylostroma extrusions.

The writer has studied the development of these xylostroma outgrowths

in cultures of both these root-rot fungi in large test tubes and jars containing lengths of woody stems or roots with the ends in agar in the bottom of the tube or jar. When the growth of the mycelial sheets between the wood and the bark becomes sufficient to cause the bark to rupture longitudinally they grow out, sometimes in frilly fashion, through the fissures thus formed. Distention of the bark by continued growth and thickening of the xylostromata results in further widening of these fissures. When freshly extruded, these xylostroma outgrowths are soft and whitish, but following exposure to air and desiccation, as occurs in nature, they soon develop a brittle, horny, dark-brown to blackish crust, like old rhizomorphs, in which grains of sand are firmly imbedded (Fig. 5). That these structures are merely

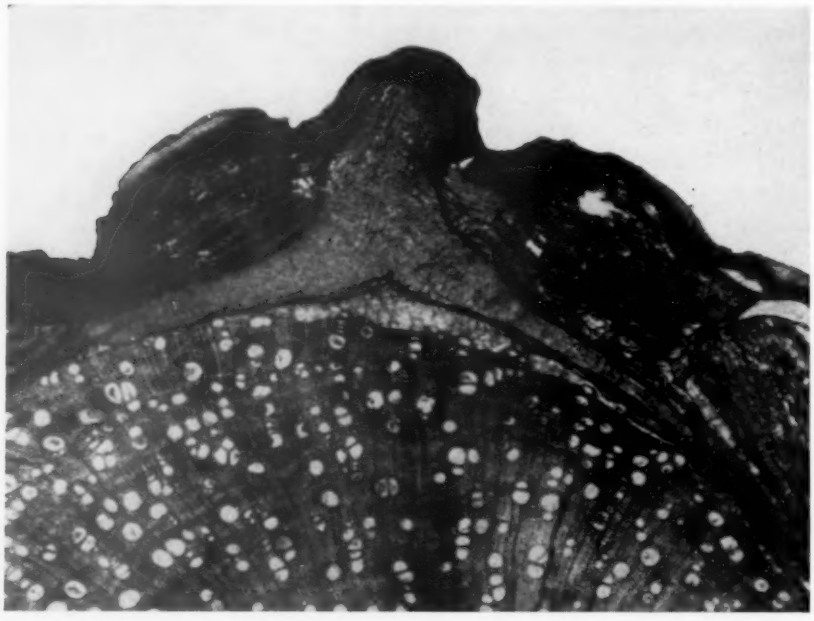

FIG. 6. Transverse section of small root of rough lemon under Marsh grapefruit attacked by Clitocybe root rot, showing xylostroma formation extruded through longitudinal fissure in bark. ×30.

outgrowths of the mycelial sheets developed between the bark and the wood, extruded through longitudinal fissures in the bark, is clearly shown by a study of transverse sections of the invaded roots (Fig. 6). These xylostroma outgrowths have been regarded as rhizomorphs by Darnell-Smith and Mackinnon (3, Fig. 2), Birmingham (1, Fig. 2) and Fawcett (4, Fig. 5, B) in their discussions of Armillaria root rot of citrus, each using the same illustrations. Although these xylostromata are comparable structurally with rhizomorphs, they are not to be confused with the usual shoestring-like rhizomorphs.

ISOLATIONS AND PATHOGENICITY OF THE CAUSAL FUNGUS

In view of the fact that *Clitocybe tabescens* has been isolated consistently

from the roots of a diverse array of plants attacked by root rot in various parts of Florida over a period of several years, only sufficient isolations were made from citrus trees to demonstrate that this fungus was consistently associated with trees attacked by root rot. From 1929 to 1935 isolations were made from the roots of 10 different living citrus trees attacked by root rot at various localities in Polk and Brevard Counties, and in each case pure cultures of *C. tabescens* were obtained. Two of these isolations were from the roots of rough lemon under grapefruit, respectively at Lake Alfred and Highland City, Polk County. Of the others, which were from Brevard County, three were from roots of rough lemon under grapefruit at Georgiana, Merritt Island, one from roots of rough lemon under calamondin at Melbourne, and three from roots of old, seedling sweet orange trees at Rockledge. All but two of these isolates developed fruiting bodies in test tube or flask cultures.

From 1924 to 1946 Clitocybe root rot has been recorded in Florida as attacking 210 different species of plants belonging to 137 genera and 59 families. The writer has isolated the causal fungus from the roots of 156 plants comprising 90 different species. These comprised native forest trees, tungoil trees, ornamental trees, shrubs and vines, and fruit trees, including banana, citrus, guava, and various other subtropical ones. Although no opportunity has been afforded to inoculate bearing citrus trees with *C. tabescens*, the repeated finding of this fungus attacking the root systems of living trees in various groves and its frequent and widespread occurrence and destructive effect on a diverse array of other trees, shrubs, and vines leaves no doubt that it is a virulent parasite capable of attacking uninjured roots of vigorously growing plants and spreading rapidly to adjacent ones, especially when closely planted. The writer (20) has shown it to be particularly destructive to plantings of beefwood (*Casuarina spp.*) trees, or so-called Australian pines, in Florida and has reported (15, 16, 17) successful inoculation experiments with it on both the horsetail beefwood (*C. equisetifolia*) and the scalybark beefwood (*C. lepidophloia*). Plakidas (6) also reported the same with young Pineapple pear trees.

RELATION OF INCIDENCE OF THE DISEASE TO SOIL CONDITIONS AND TIMBERED LANDS

Clitocybe root rot of woody plants in Florida occurs on a variety of soil types, being rather frequent on the lighter, well-drained sandy soils, which are dominantly acid in reaction and inclined to be more or less droughty. However, it also occurs on soils with an alkline reaction. In citrus trees the disease thus far has been found on but a few soil types. In the central part of the State it has been found almost exclusively on Norfolk sand, while in the few groves in Brevard County on the East Coast it occurred largely on Norfolk fine sand, though on Gainesville sand (coquina phase) in the grove at Rockledge. However, the soil type in general appears to have less influence on the incidence of the disease than has the prevalence of oak and other hardwood trees on the land prior to clearing.

The incidence of Clitocybe root rot of citrus trees in a number of groves in various parts of Polk County has been found correlated with the use of timbered land for planting. The tree growth prior to clearing was the characteristic pine-oak association, with a hammock type of growth in some cases. In practically all the 77 trees dug around for surgical treatment late in 1930 and early in 1931 in 6 groves at Lake Alfred, oak and occasionally other hardwood roots infected by *Clitocybe* were found. These citrus groves were from 9 to 19 years old and the roots that had been left in the land when it was cleared, though certainly by no means all infected at the time, enabled the fungus to spead and served to transmit it to the roots of the citrus trees with which they came into contact. It also was observed in several instances that where an infected citrus root crossed or came into contact with another citrus root that the root-rot fungus was transmitted to this root.

In the grove at Kathleen, where Clitocybe root rot was reported occurring first about 1915, it was related by the former owner that when the land was cleared in 1910 it contained numerous bluejack and turkey oaks (*Quercus cinerea* and *Q. laevis*) and that a good many stumps were left in the ground for three or more years after the trees were planted. Oak roots infected by the root-rot fungus also were found under a number of citrus trees attacked by Clitocybe root rot in one grove at Highland City. It therefore appears that oak and other hardwood roots left in the land at the time of clearing can constitute sources of infection in planted citrus trees for a greater number of years than is generally believed.

However, not all cases of Clitocybe root rot in citrus groves have been correlated with the spread of the fungus from infected roots in the land. In two groves at Georgiana on Merritt Island, where a considerable number of attacked trees were dug around for surgical treatment, oak roots were almost completely lacking, only a single small infected piece being found. However, uncleared land with oak trees adjoined the attacked trees in one grove. Moreover, in the grove at Rockledge no evidence of foreign roots was found under 4 seedling sweet orange trees, probably close to 45 years old, that were given surgical treatment. It therefore appears that infection by this disease must at times occur in some way other than by contact with infected roots, probably resulting from infection by the spores of the fungus.

COMPARATIVE SUSCEPTIBILITY AND RESISTANCE OF DIFFERENT ROOTSTOCKS

With the exception of a few old seedling sweet orange trees, an old seedling grapefruit tree, and those trees on everbearing lemon stock at Kathleen, all the citrus trees found attacked by Clitocybe root rot have been on rough lemon stock. A total of approximately 325 trees on this stock have been found attacked. Approximately two-thirds of these were budded to grapefruit, nearly one-third to orange, 9 to tangerine, 1 to calamondin, and 1 young tree to Tahiti lime.

Only in the 20-acre grove at Kathleen, which is unusual in that a number of rootstocks were used, has an opportunity been afforded to make a comparison of the incidence of Clitocybe root rot on different rootstocks in a property where this disease is known to have occurred for several years. A detailed inspection was made of this grove in June, 1935, following the receipt of the history and a diagram of the varieties and rootstocks used in the different parts, which were kindly furnished by the former owner, Mr. W. E. Tucker. Clitocybe root rot was reported to have developed about 1915 but, aside from half a dozen declining trees, the grove was said to have been in fine condition when sold in 1918. However, Mr. Tucker, who visited his home adjacent to the grove at intervals, reported that since then there had been considerable loss of trees from both root rot and the chronic wilt and decline of citrus trees known as "blight."

The trees purchased for the west three-fourths of this grove were supposed to have been budded on rough lemon stock but a considerable number actually were on everbearing lemon, as evidenced by the fruit borne on root sprouts that developed. It is well known that seed of this variety of lemon was used formerly by some nurserymen and that trees on this stock have proved short-lived and subject to "blight." Tucker stated that his trees on this stock proved unusually susceptible to both Clitocybe root rot and "blight."

In the 10-acre block of orange and grapefruit trees on the west half of this grove in which lemon (rough and everbearing) was used as the stock a large number of trees had been removed, while others had been braced with guy wires following extensive surgical treatment. As many as 17 of the original trees in one row had been either treated or removed. Immediately east of this 10-acre block were two adjoining small blocks of grapefruit trees, one being on mixed lemon stock and the other on grapefruit stock. In the former a heavy loss of trees was apparent, whereas no evidence of any tree loss occurred in the latter. This striking difference indicates a marked resistance of the grapefruit stock under conditions where the rough and everbearing lemon stocks proved highly susceptible. On the east side of this grove 4 rows of Valencia oranges on grapefruit stock were free from tree losses. In the balance of the east side of this grove, which was planted to Ruby orange on a mixture of rough lemon and sour orange stock, some losses occurred in trees on the former stock but none was apparent in those on sour orange. The complete freedom from root rot of the trees on sour orange stock in this block indicates a high degree of resistance, a characteristic that has been verified for this stock by general observations throughout a large part of the citrus region of Florida.

Although numerous groves have been examined in various sections of the State, not a single case of Clitocybe root rot has been observed on trees on sour orange stock, including the bittersweet which is botanically the same species and often used indiscriminately in mixture. No instance has been observed of citrus trees on sour orange stock being attacked by root rot,

even in groves on Merritt Island, where the disease often causes a heavy mortality of beefwood (*Casuarina spp.*) trees, or so-called Australian pines, in windbreaks planted closely around and through the groves and occasionally interplanted between the citrus trees in the same rows. Judging from these observations, trees on sour orange stock possess a high degree of resistance to Clitocybe root rot, as they do to a number of other diseases. Citrus trees, in general, appear to be much more resistant to this disease than many other woody plants.

TREATMENT OF ATTACKED TREES

It has been not only possible but quite practicable to save, by careful surgical treatment, bearing trees that are attacked by Clitocybe root rot, provided the disease be located and treatment administered early. The extent to which the disease has attacked the root system can be determined only by removing sufficient soil from around and under the root crown to permit a thorough examination. This requires removing the soil in a basin with a radius of at least from 2 to 3 feet from the trunk, working carefully to avoid injuring living roots. Unless facilities are available for washing away the soil hydraulically it should first be removed from around the base of the tree with a trowel until the location of the lateral roots can be determined. One half of a post-hole digger makes an excellent implement to remove the soil from around the root crown under the lateral roots when they are too close together to use a shovel.

If it is found practicable to treat the tree, all dead roots should be cut off flush with the root crown or living roots from which they branch, and removed in their entirety. Special effort should be made to remove any infected oak or other foreign roots that may be found under the trees being treated. All bark lesions that may occur at the base of the trunk or on the root crown, lateral roots, or taproot, in case the latter is not dead, should have the dead or infected bark cut out carefully back to healthy bark and down to sound wood, using a chisel and gouge with a mallet. The lateral roots should be inspected carefully for local lesions, particularly at their juncture with the root crown. All roots and chips of wood and bark excised should be removed and burned, because allowing such infected material to remain in the soil may lead to the redevelopment of the disease. A heavy paper placed under the exposed roots greatly facilitates the collection and removal of such material. All trees not worth treating should be dug out, and pains taken to remove and burn all roots.

After the surgical treatment, the treated areas should be brushed clean of soil and the roots inspected for any lesions that may have been overlooked. All exposed wood surfaces should then be painted with a good, durable pruning-wound compound. After this has dried, the excavation may be filled partially or completely. If the tree is not banked to stimulate the development of new roots, it is a good idea to leave the root crown ex-

posed indefinitely for aeration and drying. The soil may be disinfected with 2 to 3 gallons of a strong Bordeaux or caustic soda-Bordeaux mixture (5–5–50) poured into the soil basin. If the tops of the treated trees have died back they should be dehorned proportionately. The location of the treated trees should be mapped and they should be inspected periodically.

Frequently, the mere removal of the soil to expose the root crown and adjacent lateral roots to aeration and drying will prove very effective in checking the further development and spread of the disease on the attacked tree because the growth of the fungus is arrested by desiccation. Extensive surgical treatment of trees sometimes leads to decay of the exposed wood at the base of the trunk despite the use of a pruning-wound compound. The recovery of trees that have lost a considerable proportion of their root systems can be expedited greatly by banking or mounding the soil around the bases up to a height of several inches above the upper limit of the partial girdle to stimulate the development of new roots from the callus formation formed at the margin of the living bark. This little-known natural method of providing partially girdled trees or those deprived of many of their roots with new root systems (18) has proved of considerable value in citrus and other trees and shrubs that develop adventitious roots readily. Under favorable soil moisture conditions new roots start developing in a few months and within a year or two should attain sufficient growth to contribute materially to the support of the trees and assist in their rejuvenation.

The writer has secured very promising results from the surgical treatment of 123 citrus trees attacked by Clitocybe root rot. Success is contingent upon careful treatment and aeration of the root crown. Treatment, of course, must be administered before the disease has progressed to such an extent that the trees begin to decline. A total of 77 trees in a number of groves at Lake Alfred were treated late in 1929 and early in 1930. With the exception of one that blew over shortly afterward, all have continued to flourish and bear good crops of fruit. Thirty-six trees, including a considerable number that had begun to decline, were treated in a grove at Georgiana, Merritt Island. Of these, 20 were still living and in good condition in September, 1941; the others died and were removed at various times. Four old, seedling sweet orange trees, three of which were treated in 1931 and one in 1933 at Rockledge, and six grapefruit trees treated in 1933 at Georgiana, have all continued to thrive and were bearing good crops in September, 1941. These results show that a single surgical treatment, without soil banking in conjunction, has proved extremely effective in controlling Clitocybe root rot and in adding a considerable number of years to the life of bearing trees attacked by it in practically all cases where the trees were not too far gone when treated. Similar beneficial results have been observed in a number of trees treated surgically according to the writer's recommendations by one grower at Highland City and another at Winter Haven.

DISCUSSION

As was pointed out previously (11, 12), it has long been the custom in Florida to diagnose all decline and death of citrus trees in which the trouble was characterized by a basal girdling and death of the roots and sometimes accompanied by an exudation of gum from the bark, as foot rot (*Phytophthora parasitica*). This appears to have resulted largely from the great publicity formerly given this disease and its prevalence on seedling sweet orange trees and trees budded on this stock, which are highly susceptible to *Phytophthora*. Contrary to popular opinion, rough lemon stock has never proved particularly susceptible to foot rot. Much of the trouble that formerly was assumed to be foot rot has been shown by these investigations to be due to Clitocybe root rot instead. This disease is especially prevalent on trees on rough lemon stock and it also occurs on seedling sweet orange trees. Thus, the recognition of Clitocybe root rot and its evaluation as an important cause of tree loss in citrus groves in various parts of Florida enables us to properly diagnose this disease rather than to continue to arbitrarily assign to foot rot all diseases that girdle the trees.

SUMMARY

Mushroom root rot, caused by *Clitocybe tabescens* (Scop. ex Fr.) Bres., is discussed and evaluated as a factor of tree loss in Florida citrus groves. Although not identified as occurring on citrus trees until 1929, it is evident that this disease was active in groves several years before but that it previously was confused with foot rot, the secondary symptoms of which have a similar appearance.

A record, based on a preliminary survey, is presented of the known distribution of Clitocybe root rot on citrus trees in various counties of the State, and of isolations made of the causal fungus.

The symptoms of the disease are described in detail. Its attack usually is concentrated chiefly on the root crown and adjacent portions of the root system. It frequently results in the death of the taproot and a large proportion of the lateral roots, and sometimes causes a butt rot of the trunk before the top shows any appreciable evidence of decline. Many trees not even suspected of being attacked, but subsequently found to have their root systems invaded extensively, have been located solely by finding either fresh or dried clusters of the causal fungus fruiting at the bases. The xylostroma outgrowths frequently developing on attacked roots are described and their origin and anatomy discussed.

Clitocybe root rot occurs with greatest frequency on well-drained, and often droughty, sandy land, especially in sections where oak and other hardwood trees occurred prior to clearing. Roots of these trees attacked by the Clitocybe root-rot fungus have been found repeatedly under attacked citrus trees in groves ranging in age from 9 to 19 years. Infection in many cases results from the roots coming into contact with infected roots but the failure

to find foreign roots under trees in many cases suggests that infection may be initiated by some other means, presumably from the spores of the fungus.

Most of the citrus trees found attacked by Clitocybe root rot have been on rough lemon, with some on everbearing lemon, stock. Both are very susceptible. Seedling sweet orange trees are also susceptible, while the little used grapefruit stock appears fairly resistant and the sour orange stock very resistant. Citrus trees in general appear to be much more resistant to this disease than many other woody plants.

The treatment of attacked trees is discussed and marked success reported with surgical treatment, either alone or in conjunction with exposure of the root crowns to aeration and drying. The value of subsequently mounding the soil around the bases of treated trees to stimulate the development of new root systems is pointed out.

LITERATURE CITED

1. BIRMINGHAM, W. A. Armillaria root rot of fruit trees. N. S. Wales Dept. Agr. Plant Dis. Leaf. 18. 1920. Revised, 1934.
2. DADE, H. A. "Collar crack" of cacao. Gold Coast Dept. Agr. Bull. 5. 1927.
3. DARNELL-SMITH, G. P., and EWEN MACKINNON. Fungus and other diseases of citrus trees. Agr. Gaz. N. S. Wales **25**: 945–954. 1914.
4. FAWCETT, HOWARD S. Citrus diseases and their control. XV + 656 pp. New York. McGraw-Hill Book Co. 1936.
5. HARTIG, ROBERT. Die Zerzetzungserscheinungen des Holzes der Nadelbäume und der Eiche in forstlicher botanische und chemischer Richtung. VI + 151 pp. Berlin: Julius Springer. 1878.
6. PLAKIDAS, A. G. Infection with pure cultures of Clitocybe tabescens. Phytopath. **31**: 93–95. 1941.
7. RHOADS, ARTHUR S. Some new or little known citrus diseases observed during the past year. Proc. Florida State Hort. Soc. **43**: 80–88. Also in Citrus Industry **11**(5): 3–7, 34. 1930.
8. ———. Clitocybe root rot of grapefruit trees. U. S. Dept. Agr., Plant Dis. Reptr. **14**: 168. 1930.
9. ———. Clitocybe root rot of citrus trees and other woody plants in Florida. Citrus Industry **12**(9): 5–9, 38–40. 1931.
10. ———. Clitocybe mushroom root rot—a new disease of citrus trees. (Abstr.) Phytopath. **22**: 23. 1932.
11. ———. Clitocybe root rot of woody plants in Florida. Citrus Industry **13**(5): 11, 14. 1932.
12. ———. Unmasking new citrus root disease. Florida Grower **40**(8): 6. 1932.
13. ———. Clitocybe mushroom root rot of citrus trees and other woody plants in Florida. Florida Agr. Exp. Sta. Ann. Rept. **1932**: 142–143; **1933**: 120–121; **1934**: 76; **1935**: 91; **1936**: 89–90.
14. ———. Clitocybe mushroom root rot of woody plants. Florida Agr. Exp. Sta. Press Bull. 454. 1933; 538. 1939.
15. ———. Clitocybe mushroom root rot of citrus and other woody plants in Florida. Florida Agr. Exp. Sta. Golden Anniversary Rept. **1938**: 116–117. 1938.
16. ———. Clitocybe mushroom root rot of citrus trees and other woody plants in Florida. Florida Agr. Exp. Sta. Ann. Rept. **1939**: 121. 1939.
17. ———. Clitocybe mushroom root rot of citrus and other woody plants in Florida. Florida Agr. Exp. Sta. Ann. Rept. **1940**: 108–109. 1940.
18. ———. Growing new root systems by soil banking—a promising method of rejuvenating trees attacked by root diseases. Phytopath. **32**: 529–536. 1942.
19. ———. A comparative study of two closely related root-rot fungi, Clitocybe tabescens and Armillaria mellea. Mycologia **37**: 741–766. 1945.
20. ———. The destructiveness of Clitocybe root rot to plantings of Casuarinas in Florida. (Unpublished manuscript.)
21. ———, and E. F. DEBUSK. Clitocybe mushroom root rot. *In* Diseases of citrus in Florida. Florida Agr. Exp. Sta. Bull. 229. 1931.

THE IDENTIFICATION AND CHARACTERIZATION OF A VIRUS CAUSING MOSAIC IN MERTENSIA VIRGINICA

INEZ NIENOW[1]

(Accepted for publication September 20, 1947)

Johnson and Valleau in 1935 (10) reported a virus from *Mertensia virginica* (L.) DC. which could be transferred to Turkish tobacco producing chlorotic and necrotic rings on old inoculated leaves and a mottle similar to cucumber mosaic on new leaves. In 1946 *M. virginica* plants with mosaic symptoms were found at Urbana, Illinois. *M. virginica,* the common bluebell, is a perennial and widely distributed. This host, therefore, could serve as an important source of infection for susceptible plants. Since the cause of the disease appeared to be cucumber mosaic virus which infects several economic crops, the following group of studies was undertaken to identify and characterize the virus causing the Mertensia mosaic.

SAP INOCULATION

Inoculations were carried out with crude infectious juice from *Nicotiana glutinosa* L. and *N. tabacum* L. (Turkish). Two to four young leaves of the plant were dusted with carborundum of 600 mesh prior to being rubbed once with gauze pads saturated with infectious juice. Inoculated leaves were washed immediately to remove any toxic effects of the juice. For the quantitative studies on the effect of heat, pH, and phosphate buffer on the virus, primary leaves of blackeye cowpea which gives local necrotic lesions were used. The right half was inoculated with the variant and the left half with the control. Ten half-leaves were used for each sample. For tests on aging of the virus, whole cowpea leaves were inoculated. Local lesions were counted four days after inoculation.

INSECT INOCULATION

Aphids[2] were allowed to feed on caged *Nicotiana glutinosa* plants that were infected with Mertensia-mosaic virus. After seven days of feeding, the aphids were transferred to two healthy *N. glutinosa* plants and one *N. repanda* and allowed to feed for seven more days. These plants were screened from other insects by means of a lamp chimney covered with cheesecloth. *Myzus persicae,* after feeding on infected *Nicotiana glutinosa* plants, transmitted the virus to healthy *N. glutinosa* and *N. repanda* plants. These

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[2] Identified as *Myzus persicae* Sulzer by Dr. Herbert H. Ross of the Illinois Natural History Survey Division, Urbana, Illinois.

developed symptoms of necrosis, chlorosis, and mottling typical for the virus on these plants.

HEAT INACTIVATION

One ml. of infectious crude juice from petunia plants was placed into 3-inch serological tubes and stoppered with cotton. A control tube of 1 ml. of unheated infectious juice was prepared for each heated tube of juice. The tubes were placed in a steam-controlled agitated water bath and heated to the various desired temperatures. After a 10-minute period the tubes were quickly cooled by placing them in cold running water. All inoculating onto cowpea leaves was done at one time and within two hours after the juice was heated. The results from duplicated experiments (Table 1) indicate that the virus is inactivated somewhere between 65° C. and 70° C. for ten minutes, but that temperatures as low as 50° C. reduce infectivity.

TABLE 1.—*Effect of heat on infectivity of Mertensia-mosaic virus in crude juice*

Temperature	Mean number of local lesions on a half-leaf of cowpea	Standard deviation
50° C.	1.90	2.308
Control	7.40	4.710
55° C.	0.45	0.203
Control	8.80	6.982
60° C.	0.55	0.780
Control	8.10	5.996
65° C.	0.05	0.020
Control	10.70	7.736
70° C.	0.00	.....
Control	9.40	8.192
75° C.	0.00	.....
Control	6.70	3.471

pH AND PHOSPHATE BUFFER

Infected petunia plants were ground and the juice squeezed through four thicknesses of cheesecloth. This juice was adjusted to pH 5.5, 6.0, 7.5, 8.0, 8.5, 9.0 using KH_2PO_4 or K_2HPO_4 and 0.1 N NaOH so that the final salt concentrations were 0.1 M. phosphate. One ml. of juice was added to 9 ml. of buffer so that all data represent a 1–10 dilution of the infectious juice. Inoculations were completed within 1 hour following pH adjustments at room temperature. Concentrations of H-ions and of phosphate buffer salts influence the infectivity of tobacco-mosaic virus (16). Since 0.1 M. salt concentration was optimal (16), only this concentration of the buffers was used. From the results in table 2, it is apparent that 0.1 M. phosphate buffers increase the infectivity of the virus and that pH 6.5 is the most desirable pH for inoculation of the virus onto cowpea leaves.

AGING

Two ml. of infectious juice were placed into 3-inch serological tubes and kept at room temperature of 25°–28° C. for 1, 2, 4, 8, 12, or 48 hours prior

TABLE 2.—*Effect of pH and 0.1 M. phosphate buffers on production of local lesions on cowpea leaves by Mertensia-mosaic virus*

pH	Mean number of local lesions on a half-leaf of cowpea	Standard deviation
5.5	5.3	3.234
Control (6.7)	2.3	1.421
6.0	6.5	5.788
Control	1.7	1.519
6.5	18.2	12.569
Control	2.0	2.024
7.0	7.9	8.026
Control	2.1	2.121
7.5	2.2	1.772
Control	1.2	0.242
8.0	3.9	6.074
Control	1.6	2.061
8.5	1.3	2.439
Control	1.9	2.737
9.0	3.4	2.505
Control	2.7	3.142

to being inoculated onto the cowpea leaves. In order to evaluate the time in which it was possible to maintain optimal infectivity, the effect of aging of the virus in expressed juice was determined. The results in table 3 indicate that the host plant may influence the length of time infectious juice retains its infectivity; they also show the need for working with the virus as quickly as possible to avoid infectivity changes due to aging.

SEROLOGICAL AND CROSS PROTECTION STUDIES

In this study crude infectious juice of the petunia plant which had a consistently high concentration of virus was used as a source of antigen for eight intraperitoneal inoculations of 5 ml. each into the rabbit. Antiserum was obtained by the usual procedures. In setting up titers for flocculation tests, the antigen rather than the antiserum was diluted. Tubes were incubated for two hours at 37° C. in a water bath, and then kept overnight in the refrigerator. Initial readings were taken after incubation and final readings after refrigeration. The possibilities of serological studies as a means for plant virus identification has been demonstrated by Beale (2), Birkeland (3), Chester (4, 5, 6, 7), Gratia (8, 9), and Purdy (14, 15). Titers were

TABLE 3.—*Effect of aging at 25° C. upon the infectivity of Mertensia-mosaic virus in crude juice*

Source of virus	Total number of local lesions on four cowpea leaves, time in hours							
	0	1	2	4	8	12	24	48
Leaves, stems of petunia	17	35	31	14	7	6	0	0
Leaves of *Nicotiana sylvestris*	56	32	4	7	0	0	0	0
Leaves of *N. tabacum* (Turkish)	63	6	5	1	0	0	0	0
Leaves of *N. tabacum* (Burley)	17	52	19	23	7	.....	7	0

demonstrated against the Mertensia virus in tobacco, petunia, and cucumber as well as against *Cucumis virus*[3] in tobacco. In this study titers were obtained with absorbed antiserum of 1–128 with *Cucumis virus* in *Nicotiana glutinosa*, 1–64 with Mertensia mosaic virus in petunia, 1–32 with Mertensia-mosaic virus in cucumber. The higher titers with the *Cucumis virus* in tobacco were probably caused by a higher concentration of virus in the plant. That the virus occurring on *M. virginica* was cucumber mosaic virus was further substantiated by cross protection tests. Price (12, 13) has shown that zinnia leaves infected with a strain of cucumber-mosaic virus will not thereafter develop the necrotic lesions caused by strain 6 of cucumber-mosaic

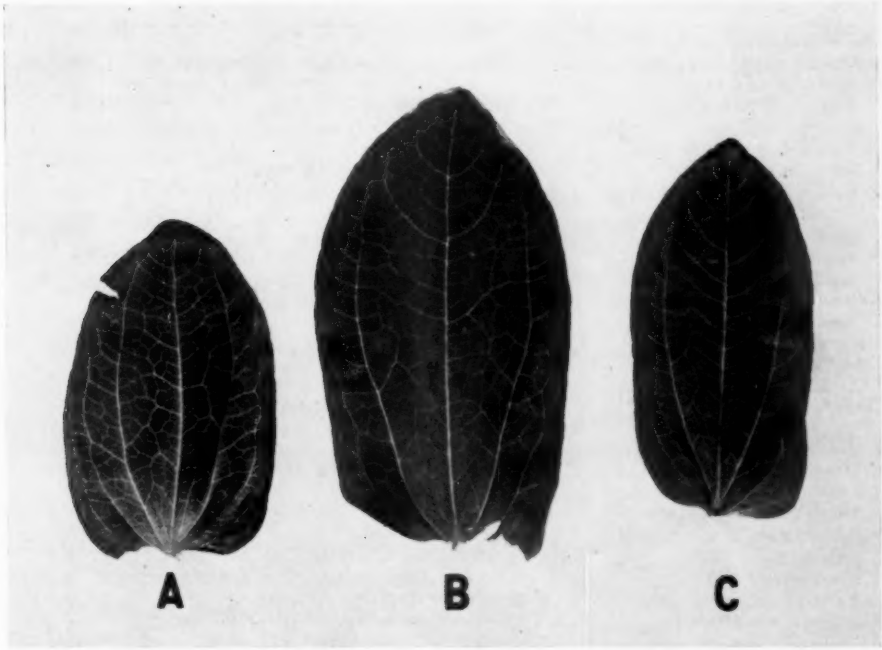

FIG. 1. Cross protection test with related strain of cucumber-mosaic virus, using strain 6 as the indicator strain which gives local necrotic lesions on zinnia leaves when the leaves have not been previously infected with a cucumber-mosaic virus: A. Infected with Mertensia-mosaic virus followed by inoculation with strain 6 cucumber-mosaic virus. B. Infected only with strain 6 cucumber-mosaic virus. C. Infected with tobacco-mosaic virus followed by inoculation with strain 6 cucumber-mosaic virus.

virus. Necrotic lesions will develop on non-infected leaves or on leaves infected with viruses other than cucumber mosaic.

When non-infected zinnia plants or zinnia plants infected with tobacco-mosaic virus were inoculated with strain 6 (indicator strain) of cucumber-mosaic virus,[4] both developed local necrotic lesions. Zinnia plants inoculated with Mertensia-mosaic virus and later with cucumber-mosaic virus strain 6 did not develop any necrotic lesions, but only showed the mosaic

[3] Obtained from Dr. S. P. Doolittle, U.S.D.A., Bureau of Plant Industry, Beltsville, Maryland.

[4] Obtained from Dr. W. C. Price, University of Pittsburgh, Pittsburgh, Pa.

from the first inoculation of Mertensia-mosaic virus (Fig. 1, A), indicating this virus is a cucumber-mosaic virus.

OVERWINTERING OF VIRUS

Diseased Mertensia plants growing in the field showed mosaic symptoms

TABLE 4.—*Summary of symptoms observed on various plants inoculated with Mertensia-mosaic virus and the results of inoculating cowpea with the juice from the respective plant species and varieties*

Species and variety of plant inoculated	Symptoms observed	Production of local lesions on cowpeas
Antirrhinum majus L.		
Golden Queen	Distortion, stunting, early flowers	+
Beta vulgaris L.		
Asgrow Wonder	Questionable	–
Early Wonder	Do	–
B. vulgaris var. *cicla* Moq.		
Spinach leaved	Systemic mottling, necrosis	+
Brassica oleracea var. *caulorapa* DC.		
Early white Vienna	No symptoms	–
B. rapa L.		
Early Purple Globe	No symptoms	–
Calendula officinalis L.		
Masterpiece	Systemic mottling	+
Cucumis sativus L.		
Cubit	Systemic mottling	+
Straight Eight	Do	+
Datura stramonium L.	Chlorotic spots on inoculated leaves, systemic mottling later	–
Delphinium ajacis L.		
Base Branching	Stunting, fern leaves, deformed flowers	+
Lactuca sativa L.		
Black Seeded Simpson	No symptoms	.....
Lycopersicon esculentum Mill.		
Prichard	Fern leaves, mottling	+
Stokesdale	Do	+
Mertensia virginica DC.	Systemic mottling	+
Nicotiana glutinosa L.	Necrosis, stunting, systemic mottling	+
N. repanda Willd.	Do	+
N. sylvestris Speg. and Comes.	Do	+
N. tabacum L.		
Burley, Ky.–16	Do	+
Burley, White	Do	+
Crittenden	Do	+
Turkish	Do	+
Petunia hybrida		
Dwarf Hybrid	Systemic mottling	+
Phaseolus vulgaris L.		
Burpee's Stringless Green Pod	No symptoms	.....
Early Golden Cluster	Do	.....
Red Kidney	Do	.....
Tendergreen	Do	.....
Rumex occidentalis S. Wats.	Do	–
Spinacia oleracea L.		
Bloomsdale Savoy	Necrosis, stunting	+
Tagetes patula L. (Hybrid)		
French Marigold	Necrosis of stem and petiole	+
Vigna sinensis Endl.		
California Blackeye	Primary necrotic lesions	+
Blackeye	Do	+
Zinnia elegans Jacq.	Mottling, distortion	+

the following spring. This indicated that the virus persisted in the perennial tissues of the plant during the winter months and appeared, subsequently, in the above ground tissues where the virus became subject to spread by feeding insects to additional plants.

SYMPTOMS PRODUCED BY MERTENSIA MOSAIC VIRUS

As one means of identification, the virus was inoculated into a wide range of plants in order to compare the symptoms produced with symptoms of other viruses in these hosts. The results are summarized in table 4.

INCLUSION BODIES

Epidermis from infected leaves, stems, and petioles was stripped from infected *Nicotiana tabacum* (Turkish) and *N. glutinosa* and stained for plant virus inclusion bodies according to the methods used by McWhorter (11) with modifications suggested by him.[5] No inclusion bodies were found, which suggested that the virus is one that does not produce inclusion bodies. According to Bawden (1), cucumber viruses 1, 3, 4, and the viruses of potato leaf roll, potato paracrinkle, tomato spotted wilt, and tomato big bud do not produce intra-cellular inclusions.

SEED TRANSMISSION

Seeds obtained from infected *Nicotiana glutinosa, N. repanda,* and *N. tabacum* (Turkish) were sown and the seedlings examined for evidence whether the virus was seed transmitted. Among approximately 100–150 seedlings of each of the three kinds of tobacco, no diseased plants developed.

DISCUSSION

In the early studies, during September through December, 1946, it was difficult to transmit Mertensia-mosaic virus to plants of any variety or species because the temperature of the greenhouse fluctuated from 60° to 75° F. When the plants were moved to a greenhouse in which the temperature was maintained at 70°–74° F., symptoms of infection readily appeared on a wide range of hosts. It was noted that the virus affected the flowers of all plants which became infected: the flower buds failed to mature, fell off, or produced flowers that were small and distorted. The concentration of virus in plants, tested by the number of local lesions produced on primary leaves of the cowpea, varied considerably, *e.g.*, it was low in snapdragon and calendula, but high in petunia and some varieties of tobacco. Young plants are preferable for such studies since old plants often showed marked effects of disease yet virus concentration was not proportionately high.

Before conducting any quantitative studies, infectious plant juice was tested for concentration by inoculation onto cowpea leaves in order to give some assurance of sufficient local lesions to make such studies valid. It was apparent from the results that the Mertensia-mosaic virus rapidly lost its

[5] Personal communication dated February 14, 1947.

infectivity upon aging and that the ability to withstand aging varied with the host plant, the age of the plant, and possibly other undetermined factors. The Mertensia-mosaic virus was rather sensitive to pH changes, and best infections were obtained between pH 6.5 and 7.0. This pH was about that of the petunia plant but whether this was a factor in the production of high concentration of virus in this plant is conjecture.

Phosphate buffers at 0.1 M. concentration increased the number of local lesions produced. The phosphate buffer may have released more virus from particulate material of the crude juice, or dispersed virus aggregates by increasing the solubility, or have affected the cells of the plant tissue in such a manner that infection was increased.

Since the Mertensia-mosaic virus was readily adsorbed, amenable to aging, and destroyed by freezing, it was not practical to filter or freeze the antigen before injection. Crude, freshly extracted plant material injected immediately gave the best results. It is advisable to determine relative amounts of virus in antigenic material that is to be used for injection into animals, since this gives assurance of an antiserum of high titer.

SUMMARY

1. The virus causing mosaic in *Mertensia virginica* (L.)DC. was identified by serological tests, cross protection tests, host range, and insect transmission as *Cucumis virus* I Smith.
2. The virus was transmissible to a large number of plants and produced a variety of symptoms dependent on the host plant.
3. The virus was inactivated between 65° and 70° C. for ten minutes.
4. Infectivity of the virus was increased by 0.1 M. phosphate buffer.
5. The most desirable H-ion concentration for inoculation on cowpea leaves was pH 6.5.
6. It was expedient to work with the virus within a two-hour period to eliminate the decrease in infectivity due to aging.
7. The virus was not transmitted by seeds of infected *Nicotiana repanda*, *N. glutinosa* L., *N. tabacum* L. (Turkish).
8. The virus was transmitted by *Myzus persicae* Sulzer.
9. *Mertensia virginica* (L.)DC. is a new host for *Cucumis virus* I and an important virus reservoir since it is a perennial and the virus persists in the host.

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LITERATURE CITED

1. BAWDEN, F. C. Plant viruses and virus diseases. Ed. 2. 294 pp. Waltham, Mass. 1943.
2. BEALE, H. A. (PURDY). Specificity of the precipitin reaction in tobacco mosaic disease. Contr. Boyce Thompson Inst. 3: 529–539. 1931.
3. BIRKELAND, JORGEN M. Further serological studies of plant viruses. Ann. Appl. Biol. 22: 719–727. 1935.
4. CHESTER, K. STARR. Serological evidence in plant-virus classification. Phytopath. 25: 686–701. 1935.

5. ————————. Separation and analysis of virus strains by means of precipitin tests. Phytopath. **26**: 778–785. 1936.
6. ————————. Serological studies of plant viruses. Phytopath. **27**: 903–912. 1937.
7. ————————. A simple and rapid method for identifying plant viruses in the field. Phytopath. **27**: 722–727. 1937.
8. GRATIA, A. Pluralité antigénique et identification sérologique des virus des plantes. Compt. rendu Soc. Biol. **114**: 923–924. 1933.
9. ————————. Qualité antigénique des virus des plantes et des bacteriophages. Compt. rendu Soc. Biol. **114**: 925–926. 1933.
10. JOHNSON, E. M., and W. D. VALLEAU. The ring symptom of virus diseases of plants. Ky. Agr. Exp. Sta. Res. Bull. 361: 239–263. 1935.
11. MCWHORTER, FRANK P. Plant virus differentiation by Trypan-blue reactions within infected tissue. Stain Technology **16**: 143–148. 1943.
12. PRICE, W. C. Acquired immunity from cucumber mosaic in zinnia. Phytopath. **25**: 776–789. 1935.
13. ————————. Cross protection tests with two strains of cucumber-mosaic virus. Phytopath. **29**: 903–905. 1939.
14. PURDY, H. A. Immunological reactions with tobacco mosaic virus. Proc. Soc. Exp. Biol. Med. **25**: 702–703. 1928.
15. ————————. Immunological reactions with tobacco mosaic virus. Jour. Exp. Med. **49**: 919–935. 1929.
16. THORNBERRY, H. H. Effect of phosphate buffers on infectivity of tobacco-mosaic virus. Phytopath. **25**: 618–627. 1935.

PRODUCTION OF ANTIBIOTIC SUBSTANCES BY FUSARIA[1, 2]

DIEGO A. TEXERA[3]

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HISTORICAL AND INTRODUCTORY WORK

The observation that molds of the genus *Fusarium* are able to excrete into artificial substrates metabolic products toxic to other microorganisms dates back to 1924 when Boyle and Pratt (4, 14) performed their classical studies on staling of *Fusarium* cultures.

In the field of plant pathology the antagonistic properties of fusaria have been investigated from two main angles: studies on toxic excretory substances capable of producing wilts in plants (6, 8, 13, 16, 24) and observations about their antifungal power (15, 23, 26). None of these studies was concerned with the isolation of true antibiotics of possible use in chemotherapy.

In 1937 Ashley, Raistrick, and their coworkers (3) isolated two quinone pigments, rubrofusarin and aurofusarin, and one non-pigmented substance, culmorin, from pure cultures of *Fusarium culmorum*. As far as we know, these substances have never been studied from the standpoint of antibiotics.

Still another pigment isolated from a species of *Fusarium* is javanicin, recently announced by Arnstein, *et al.* (1, 2), from *F. javanicum*. Unlike rubrofusarin and the others, the antibiotic properties of this substance have been thoroughly investigated.

Aside from this work, there have been in the past few years occasional reports concerning the ability of fusaria to inhibit the growth of other microorganisms, particularly bacteria (5, 7, 20, 25), but most of these observations have consisted of brief references incidental to extensive surveys in which the antibacterial effect of hundreds of fungal cultures was examined.

The present study is concerned with the isolation of two antibiotic substances from *Fusarium hyperoxysporum* Woll., their mode of formation, and their properties.

COMPARISON OF SAPROPHYTIC AND PLANT PATHOGENIC FUSARIA[4] FOR BACTERIOSTATIC ACTIVITY

The introductory phase of the investigation consisted in the isolation of eighteen *Fusarium* strains from soil and composts and the examination of their bacteriostatic activity in different liquid culture media after preliminary screening by means of the cross-streak test (19). The artificial substrates compared were:

[1] Journal Series paper of the New Jersey Agricultural Experiment Station, Department of Microbiology, Rutgers University.

[2] The author is grateful to Dr. Selman A. Waksman for suggesting the problem and for his valuable criticism during the progress of the work.

[3] Present address: Instituto Experimental de Agricultura, Caracas, Venezuela.

[4] The author is indebted to Dr. C. M. Haenseler, of the Plant Pathology Department, for the cultures of the plant pathogenic fusaria used in this work.

1. Glucose-peptone.
2. Glucose-asparagine (Krainsky's).
3. Glucose-nitrate (Czapek-Dox).
4. Glucose-nitrate with 0.5 per cent corn steep liquor.
5. Sucrose-nitrate (brown sugar medium).

The culture media were dispensed in 50-ml. volumes in Erlenmeyer flasks of 250-ml. capacity, neutralized, and sterilized at 10 lb. steam pressure during one-half hour. Inoculation was made with one-ml. portions of spore suspensions prepared from one-week-old slants of the strains reared in glucose-peptone agar. Incubation followed at 28° C. for two weeks and the

TABLE 1.—*Antibiotic activity of saprophytic and plant pathogenic strains of Fusarium grown in glucose-nitrate medium*

Strain	Dilution units per 1.0 ml. at 14 days of incubation							
	Metabolic solution[a]				Mycelium[b]			
	E. coli	*S. aureus*	*B. subtilis*	*B. mycoides*	*E. coli*	*S. aureus*	*B. subtilis*	*B. mycoides*
Fusarium sp. 21	< 5	< 5	90	90	< 30	400	1,000	2,000
Fusarium sp. 25	< 5	< 5	80	150	< 30	125	450	450
Fusarium sp. 26	< 5	< 5	50	150	< 30	125	1,250	1,250
Fusarium sp. 30	< 5	< 5	50	50	< 30	400	1,250	1,250
Fusarium sp. 31	< 5	< 5	500	500	< 30	250	1,250	1,250
Fusarium sp. 32	< 5	< 5	< 5	< 5	< 30	< 30	< 30	30
Fusarium sp. 37	< 5	< 5	< 5	< 5	< 30	30	50	100
Fusarium sp. (from asparagus)	< 5	< 5	200	200	< 30	50	200	500
F. bulbigena (from tomato)	< 5	< 5	80	90	< 30	50	150	250
F. apii (from celery)	< 5	< 5	< 5	< 5	< 30	30	50	50
F. hyperoxysporum (from sweet potato)	< 5	150	600	1,000	< 30	500	1,000	2,500

[a] Final pH of all cultures: 6.6–6.8.
[b] Each pellicle was extracted with 50 ml. of 95 per cent ethyl alcohol.

tests for antibiotic activity were performed at one and at two weeks of age on both the culture filtrates and the mycelium. This consisted in draining the pellicles and extracting them in 50 ml. of 95 per cent ethyl alcohol overnight; the antibiotic tests were performed next day on the filtered alcoholic extracts. Previous experience had shown that methyl alcohol and acetone were as effective as ethyl alcohol for the extraction of the antagonistic agent from the mycelium. The antibiotic activity was determined by means of the agar dilution method (21) and the figures express dilution units per 1.0 ml. of test material. Since little or no difference in antibiotic activity was observed between the one- and two-week-old cultures, only one set of figures is given.

By experiment it was determined that 0.33 ml. of alcohol is the largest

amount of this solvent which will allow normal growth of the test organisms; therefore the 30 dilution units was the lowest level of activity detectable for the mycelial extracts.

This experiment yielded the following information:

(1) Of the 18 soil isolates tested, 6 were capable of producing a bacteriostatic principle and this was more pronounced in the glucose-nitrate medium without reinforcements; the addition of 0.5 per cent corn steep liquor completely suppressed the formation of the substance. Glucose-asparagine and glucose-peptone media produced very low antibacterial activity.

(2) The antibiotic substance was more abundant in the mycelium than in the metabolic solution, which shows that it is produced intracellularly and excreted in varying degree into the substrate.

(3) The antagonistic principle was active against the Gram-positive but not against the Gram-negative test organisms used. The usefulness of

TABLE 2.—*Vegetative degeneration and shift in bacteriostatic spectrum of Fusarium hyperoxysporum in glucose-nitrate medium*

Age in days	Mg. residual sugar per ml.	pH	Dilution units per ml. of culture filtrate			
			S. aureus	*B. subtilis*	*B. mycoides*	*E. coli*
0	38.81	6.7	.....	.....	.....	.....
3	32.56	6.2	< 5	< 5	< 5	< 5
5	27.61	5.9	< 5	< 5	< 5	< 5
7	22.82	5.7	10	30	30	< 5
10	20.15	6.1	30	65	.....	.....
13	17.40	6.0	65	100	65	20
17	11.00	7.1	100	300	350	30
20	5.35	7.2	100	250	250	25

glucose-nitrate medium for the production of antagonistic activity by the *Fusarium* strains having been determined, twenty-one new soil and manure isolates were tested against 4 plant pathogenic strains. The extremely high order of bacteriostatic activity of some of these isolates is shown in table 1.

ANTAGONISTIC MODIFICATION OF ACTIVE STRAINS

Upon transference of the experimental work to Louisville, Kentucky,[5] the active saprophytic and plant pathogenic fusaria suffered three important modifications, namely:

(1) Marked loss of vegetative vigor.

(2) Shift in bacteriostatic spectrum demonstrable by the fact that Gram-negative organisms began to be inhibited in addition to the Gram-positives.

(3) Eventual loss of the antagonistic power.

Table 2 illustrates the new behavior of *Fusarium hyperoxysporum* before its bacteriostatic ability was completely lost. The loss of vigor was evident by the slow growth made and by the fact that thick and convoluted pellicles

[5] Acknowledgment is made of the facilities accorded by the firm of Joseph E. Seagram & Sons of Louisville, Ky., during part of this work.

were no longer formed. Besides these visible characters, the cultures failed to utilize the sugar completely and the residual pH of the medium did not rise. The alcoholic extracts of the scant mycelium obtained showed very low antagonistic activity.

VEGETATIVE REACTIVATION OF STRAINS

It was soon found that the poor growth made by the strains was due to an insufficiency of zinc in the tap water. It is well to recall in this connection that the formula of the glucose-nitrate (Czapek-Dox) medium calls for 10 p.p.m. ferrous sulphate but for no added zinc. When this element was added in Louisville in the proportion of 10 mg. of zinc sulphate per liter of medium, vigorous growth resulted in one week, the sugar was completely and rapidly utilized, and the pH of the metabolic solution rose to 8.5 or more. Where zinc was added the resulting pellicles were thicker, raised, and wrinkled, while the remaining treatments produced thinner and lighter pads with no convolutions.

This restoration of vegetative vigor was not accompanied, however, by the expected recovery of the antagonistic power. The beneficial effect of zinc on growth is in line with the findings of other investigators (9, 10, 11, 17) for molds of the genera *Rhizopus*, *Aspergillus*, and *Penicillium*.

ANTAGONISTIC REACTIVATION OF FUSARIUM HYPEROXYSPORUM. TWO-MEMBERED CULTIVATION WITH BACTERIA AND SPORE SELECTION

It has been shown elsewhere (22) that the enrichment of normal soils with live and washed *Escherichia coli* cells brings about a decided stimulation in the number of fusaria in the soil, and it is well to mention that advantage was taken of this phenomenon for the isolation of saprophytic strains in the preliminary phase of the present work. None of the isolates, however, had been able to inhibit the growth of *E. coli*, although this bacterial species was inhibited later and for the first time in Louisville by a plant pathogenic *Fusarium*. In both cases, nevertheless, antagonistic capacity was eventually lost.

In an attempt to study in more detail this stimulative effect of bacterial enrichment upon fusaria and its possible role upon their antagonistic behavior, two-membered cultures of *Fusarium hyperoxysporum* with *Escherichia coli* and with *Bacillus mycoides* were prepared in sterilized soil accompanied by a pure culture of the fungus in soil alone. The cultures were incubated at 28° C. and kept moist by periodic irrigations with sterile water. Microscopic examination and periodic platings into glucose-peptone and nutrient agar showed that the organisms had been firmly established in the soil mass. Constant watch was maintained on their purity.

After one month of incubation a series of platings was made from the pure and the two-membered cultures on acidified glucose-nitrate agar, and individual colonies were transferred into duplicate slants of the same

medium. Parallel platings were made from the regular stock culture, which, when propagated directly by means of mass transfer, consistently yielded growth with very little or with no antagonistic capacity. This stock culture had been maintained in soil-extract agar (12) because it had been previously found that this substrate produced growth with more raised mycelium. Five colonies from each substrate were propagated using one member of each duplicate.

The propagation was made by the usual technique of suspending the growth from the agar slants in a small volume of sterile water or saline, and inoculating one ml. of the suspensions in duplicate into 50-ml. volumes of glucose-nitrate broth of pH 7. This time the medium was reinforced with 10 p.p.m. of zinc sulphate to ensure vigorous growth. The 40 cultures were incubated at 28° C. and tested at 1 and at 2 weeks of age for antibiotic activity against *Bacillus mycoides* and *Escherichia coli* by the agar dilution method. One member of each pair was used each time and both the metabolic solutions and the alcoholic extracts of the mycelium were tested. Since

TABLE 3.—*Antibiotic activity of colonies of Fusarium hyperoxysporum obtained by plating 2-membered and pure cultures from different substrates*

Substrate	*B. mycoides* dilution units per ml. of alcoholic extract of mycelium[a]				
Soil	50	< 30	< 30	< 30	50
Soil and *E. coli*	300	< 30	650	300	< 30
Soil and *B. mycoides*	< 30	< 30	100	< 30	150
Soil extract agar	< 30	400	300	300	< 30

[a] Each pellicle was produced in 50 ml. of medium in a 250-ml. Erlenmeyer flask and was extracted with 50 ml. of 95 per cent ethyl alcohol.

the antibiotic level of the solutions was almost negligible, only the results of the mycelial extracts are recorded in table 3. The antagonistic titer remained unchanged after the first week of incubation. No inhibition could be recorded against *E. coli* in any of the cultures.

The most striking result of this experiment was the manifest ability of spores proceeding from the degenerated stock culture to produce antagonistic pellicles. This was certainly a very unexpected result in view of the consecutive number of inactive and nearly inactive cultures previously obtained from it by mass transfer.

In regard to the very important point of the influence of *Escherichia coli* upon the antagonistic behavior of the fungus, the results were negative. This agrees with the results obtained by Waksman and Schatz (22) with normal soil cultures in which the increased number of fusaria obtained by enrichment with *E. coli* failed to inhibit this Gram-negative organism although antagonizing considerably the Gram-positives.

The spores previously harvested from the highest testing pellicle of table 3 were plated out and 20 of the resulting colonies propagated and tested. The process was repeated twice more, the previously harvested spores of

the highest testing pellicle of each generation always being plated. The results of this work can be summarized thus:

(1) No increase above the 650 *Bacillus mycoides* dilution units level was obtained.

(2) Inactive pellicles were eliminated by selection.

(3) The general level of activity was improved in the first two generations but receded again in the third.

(4) Not one culture inhibited *Escherichia coli.*

EFFECT OF ZINC ON THE BACTERIOSTATIC SPECTRUM OF FUSARIUM HYPEROXYSPORUM

It has been shown that when the antagonistic reactivation of the fungus was accomplished in Louisville through vegetative segregation, the antibacterial spectrum obtained conformed, not to the one observed there before the mold lost its activity, but to the original pattern of inhibition previously obtained in New Brunswick. Moreover, the fact that the reactivation had been registered in glucose-nitrate medium reinforced with zinc (in order to ensure vigorous growth) strongly suggested that perhaps the absence of this element might have been responsible for the shift in antibacterial spectrum already noted.

This possibility was investigated upon resumption of the work in New Brunswick. The study consisted in observing again the antibacterial behavior of *Fusarium hyperoxysporum* grown in glucose-nitrate medium with different levels of zinc and other metals and using a wider range of test bacteria. In order to refine the conditions, the water used for the preparation of the medium was freed from trace elements by the Steinberg technique (18), which consists in thoroughly mixing the medium with 2.5 per cent calcium carbonate followed by heating under steam pressure and filtering. All glassware was rinsed with this purified water. The chemicals used in the preparation of the media were of Merck Reagent grade, except the glucose, for which commercial "cerelose" was used. The basal medium consisted of the original Czapek-Dox formula, which includes 10 p.p.m. added ferrous sulphate but no zinc.

It was possible to show that when antagonistically active cultures of *Fusarium hyperoxysporum* are grown in glucose-nitrate medium prepared with ordinary tap water or with purified distilled water reinforced with zinc, the antibiotic principle produced is active against Gram-positive bacteria only. When the medium is prepared with distilled water purified by the Steinberg method, however, the antibiotic substance produced can inhibit both the Gram-positive and the Gram-negative organisms.

Likewise, under conditions of zinc deficiency, the active principle is readily excreted into the metabolic solution in contrast with the ordinary zinc-satisfied cultures in which the activity remains tied up in the mycelium. It was likewise observed that the zinc-deficient growth produced higher levels of activity in submerged cultivation obtained with a rotary shaking

machine. These cultures produced scant mycelium, failed to consume all the sugar, and their hydrogen ion concentration remained close to neutrality, which was precisely the behavior observed in Louisville before the medium was reinforced with zinc. Table 4 illustrates a typical experiment. Further information obtained in this study can be summarized thus:

(1) The zinc effect takes place in the presence as well as in the absence of iron.

(2) Manganese and copper do not exert the slightest effect on the behavior of the fungus.

(3) Individual spore isolates, although showing marked quantitative variability, behave in exactly the same way qualitatively toward the metal.

TABLE 4.—*Effect of zinc upon the bacteriostatic spectrum of Fusarium hyperoxysporum*

Test bacteria	Dilution units per ml. after 14 days of incubation	
	Stationary cultures[a]	Shaken cultures[b]
	Alcoholic extract of mycelium; medium with 10 p. p. m. zinc sulphate added	Metabolic solutions of zinc-free medium
Bacillus mycoides	500	90
B. subtilis	500	150
Staphylococcus aureus	200	35
Escherichia coli	< 30	40
Shigella paradysenteriae	< 30	150
Salmonella pullorum	< 50	90
Proteus vulgaris	< 30	50

[a] Produced in 50 ml. of medium in 250-ml. Erlenmeyer flasks.
[b] Produced in 100 ml. of medium in 250-ml. Erlenmeyer flasks.

Because of their difference in antibacterial spectrum and in mode and characteristics of formation, the two antibiotic substances produced by *Fusarium hyperoxysporum* were regarded as distinct from one another and tentatively were designated as A and B.

Thus fraction A stands for the substance produced in the presence of zinc, active mainly against the Gram-positive bacteria, and obtainable only from the mycelium of the fungus in its present level of antagonistic activity.

Fraction B designates the substance produced in the absence of zinc, active against both Gram-positive and Gram-negative bacteria and obtainable from the metabolic solution.

EXTRACTION OF FRACTION A FROM MYCELIUM

The fungus was cultivated in 10 liters of glucose-nitrate medium prepared with tap water and reinforced with 5 p.p.m. of zinc sulphate to ensure against any deficiency of this element. After two weeks' incubation at 28° C. in stationary culture, the pellicles were separated from the medium, squeezed through cheesecloth, and extracted with three consecutive half-

liter volumes of 95 per cent ethyl alcohol for 24 hours each time. After pooling and filtering, a volume of 1,675 ml. of a golden brown extract was obtained.

Previous experience had demonstrated that this alcoholic extract would not yield any of its antibacterial activity to Norit A, a powdered charcoal adsorbent, when undiluted, but that, upon dilution with an equal volume of distilled water, more than 90 per cent of the activity could be adsorbed in 1 per cent Norit based on the final volume. The antibiotic test made on the final 3,350 ml. of dilute extract gave the following values in dilution units per ml.:

Bacillus mycoides	750	*Shigella paradysenteriae*	< 30
Staphylococcus aureus	300	*Salmonella pullorum*	< 50
Escherichia coli	< 30	*Proteus vulgaris*	< 30

The liquid was then treated with 1 per cent Norit A with mechanical stirring for one-half hour; the antibiotic test performed with the filtrate showed 25 dilution units for *Bacillus mycoides* and no detectable action for *Staphylococcus aureus.*

Previous experience had shown that, once the active principle is adsorbed in the Norit, it cannot be eluted with neutral or acid alcohol but passes readily into alkaline alcohol prepared by mixing 11 parts of 95 per cent ethyl alcohol with 4 parts of distilled water alkalinized with 2 per cent NaOH based on the final volume. Because of this experience the Norit was eluted three times in succession with 2 per cent alkaline alcohol at room temperature for two hours each time and with frequent shaking. Pooling of the eluates gave a volume of 845 ml. with a titer of 1,200 *Bacillus mycoides* dilution units per ml. after neutralization with concentrated HCl. This is equivalent to 40 per cent recovery for this test organism.

After concentration of the eluates to 200 ml. under diminished pressure, the resulting cloudy suspension was slowly added with constant stirring to 2 liters of acetone, the salts were filtered off, and the acetone was removed by distillation. After it was ascertained that more than 90 per cent of the antibiotic activity was in the organic fraction, this was carefully collected, made up to 200 ml. with distilled water, and extracted twice with four volumes of diethyl ether with vigorous shaking. Evaporation of the ether layers produced 500 mg. of a brown, viscous residue with a pungent odor and which could not be dried under vacuum. The antibiotic spectrum obtained with the alcoholic solution of this crude preparation is given in table 5. It can be seen that this spectrum bears no resemblance to the antibacterial spectra of penicillin, chaetomin, or fumigacin (19).

ISOLATION OF FRACTION B FROM METABOLIC SOLUTION

Ten liters of glucose-nitrate medium were prepared with distilled water purified by the Steinberg technique already described and inoculated with spores of *Fusarium hyperoxysporum.* Because the incubation was done in

stationary culture the antibacterial titer of the metabolic solution remained very low even after 20 days at 28° C. The growth of the fungus was very poor in this zincless medium and the pH did not rise above 7.2 After separation from the mycelium and filtration, 8.0 liters of metabolic residue were obtained. The antibiotic strength of this solution, measured in dilution units per ml. was as follows:

Bacillus mycoides	25	*Proteus vulgaris*	5
Escherichia coli	5	*Shigella paradysenteriae*	20

The active filtrate was now treated with 80 grams of Darco activated carbon (grade G–60) by mixing at room temperature for one hour with

TABLE 5.—*Antibiotic spectra of Fusarium fractions A and B*

Test organism	Dilution units per 1.0 mg. dry weight of final alcoholic solution	
	Fraction A	Fraction B
Staphylococcus aureus (ATCC 6538)	117	20
Sarcina lutea No. 14	290	10
Micrococcus lysodeikticus No. 19	190	20
Bacillus cereus No. 8	784	40
B. subtilis (ATCC 6633)	686	40
B. mycoides (ATCC 6462)	585	35
B. megatherium No. 10	190	35
B. circulans	190	35
Mycobacterium phlei No. 23	137	15
M. tuberculosis var. *hominis* (ATCC 607)	30	5
Escherichia coli No. 4	< 10	15
Proteus vulgaris No. 73	30	15
Proteus vulgaris No. 74	30	15
Serratia marcescens (ATCC 989)	< 10	5
Salmonella pullorum (PCI No. 904)	< 10	35
Shigella paradysenteriae ("V"9380–ATCC)	< 10	35
Aerobacter aerogenes No. 66	< 10	< 5
Pseudomonas fluorescens No. 68	< 10	< 5
Candida albicans No. 173	15	15
Trichophyton mentagrophytes No. 171	30	10
Trichoderma sp. No. 102	15	< 5
Fusarium hyperoxysporum Woll.	< 10	< 5

mechanical stirring. After overnight standing in the refrigerator the carbon was filtered off and dried at 55° C. overnight followed by 24 hours at room temperature. The dried adsorbent was then treated with 400 ml. of diethyl ether for 24 hours followed by elution with 800 ml. of 95 per cent ethyl alcohol overnight at room temperature. This preliminary treatment with ether was adopted because previous experience had shown that it facilitated the removal of the antagonistic substance by the alcohol. The alcohol eluate was then filtered and evaporated under diminished pressure to a very small volume followed by overnight drying at 55° C. in the dry oven. The complete evaporation of the alcohol and water resulted in a light brown and transparent brittle residue which was then redissolved in 50 ml. of warm 95

per cent alcohol. Treatment of this alcoholic concentrate with ten volumes of cold distilled water produced precipitation of a white crystalline fraction. Two more precipitations were obtained by evaporating the mother liquor under diminished pressure, redissolving the residue in alcohol, and treating again with water; 1.7 grams of dried crystalline preparation was obtained from 8 liters of metabolic solution. The antibiotic spectrum of this substance is summarized in table 5; it bears some resemblance to the spectrum of penicillic acid (19).

BEHAVIOR OF FRACTIONS A AND B "IN VIVO"[6]

Toxicity and efficacy tests conducted with samples of the two substances showed that fraction A is nontoxic when administered subcutaneously in concentrations of 125 *Staphylococcus aureus* dilution units per mouse. At this concentration, however, it failed to protect the animals against infection with *S. aureus SM*. This suggests inactivation or destruction of the substance in the animal organism.

The *in vivo* work with fraction B was complicated by the low antibiotic potency of the crystalline preparation, which required the administration of 5.0 mg. per mouse in order to bring the antibiotic unitage to the minimum level necessary for the efficacy tests. This dose was found to be toxic in subcutaneous injection. The efficacy tests showed that when doses just below the toxic level were administered the substance failed to protect the experimental animals against *Salmonella schottmülleri*. It might be possible that further purification of this antibiotic can increase its potency to a point at which protection would be feasible with doses below the toxic level.

SUMMARY

When several saprophytic and plant pathogenic fusaria were propagated in simple synthetic media like glucose-nitrate (Czapek-Dox) broth, they readily produced antibiotic substances. This property was quickly lost, however, when the stock cultures were perpetuated by mass transfer but was regained through the propagation of individual spores.

Further work with *Fusarium hyperoxysporum* demonstrated that the antagonistic potency of this organism against the Gram-positive bacteria could be enhanced to some degree by two-membered cultivation with *Escherichia coli* in sterile soil. This association, however, did not result in the inhibition of *E. coli* by the fungus.

The qualitative antagonistic behavior of *Fusarium hyperoxysporum* was found to be governed by the quantity of zinc present in the culture medium. In the presence of this element the fungus produces an antibiotic active mainly against Gram-positive bacteria; in its absence both the Gram-positive and Gram-negative organisms are inhibited.

Both antibiotic principles were isolated from mass cultures of *Fu-*

[6] The author is indebted to Mr. Otto E. Graessle of the Merck Institute for Therapeutic Research of Rahway, N. J., for this information.

sarium hyperoxysporum and designated tentatively as A and B. Further chemical work is needed to identify and characterize these substances. Fraction A proved to be nontoxic to experimental animals but failed to convey protection against infection with *Staphylococcus aureus SM.* Fraction B was toxic in doses of 5.0 mg. per mouse, and when administered in amounts below the toxic level it did not protect the animals against *Salmonella schottmülleri.*

In its mode of formation and in antibacterial spectrum, fraction A is distinct from penicillin, chaetomin, and fumigacin, while fraction B bears some resemblance to penicillic acid. Further work is needed to elucidate the true nature of these active principles.

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LITERATURE CITED

1. ARNSTEIN, H. R. V., A. H. COOK, and M. S. LACEY. An antibacterial pigment from *Fusarium javanicum.* Nature **157**: 333–334. 1946.
2. ————, ————, ————. Production of antibiotics by fungi. Part II. Production by *Fusarium javanicum* and other fusaria. Brit. Jour. Exp. Path. **27**: 349–355. 1946.
3. ASHLEY, J. N., B. C. HOBBS, and H. RAISTRICK. LV. Studies in the biochemistry of microorganisms. LIII. The crystalline colouring matters of *Fusarium culmorum* (W. G. Smith) Sacc. and related forms. Biochem. Jour. **31**(I): 385–397. 1937.
4. BOYLE, C. Studies in the physiology of parasitism; the growth reactions of certain fungi to their staling products. Ann. Bot. **38**: 113–135. 1924.
5. BOISSAVAIN, C. H. Growth inhibition of tubercle bacilli by *Fusarium* sp. Proc. Soc. Exp. Biol. and Med. **63**: 555–556. 1946.
6. BRANDES, E. W. Banana wilt. Phytopath. **9**: 339–389. 1919.
7. CARPENTER, C. W. Antibacterial properties of yeasts, *Fusarium* sp., onion, and garlic. Hawaii Plant Rec. **49**(I): 41–67. 1945.
8. FAHMY, T. The production by *Fusarium solani* of a toxic excretory substance capable of causing wilting in plants. Phytopath. **13**: 543–550. 1923.
9. FOSTER, J. W., and S. A. WAKSMAN. The specific effect of zinc and other heavy metals on growth and fumaric acid production by Rhizopus. Jour. Bact. **37**: 599–617. 1939.
10. ————, H. B. WOODRUFF, and L. E. MCDANIEL. Microbiological aspects of penicillin. III. Production of penicillin in surface cultures of *Penicillium notatum.* Jour. Bact. **46**: 421–433. 1943.
11. ————, ————, and ————. Microbiological aspects of penicillin. IV. Production of penicillin in submerged cultures of *Penicillium notatum.* Jour. Bact. **51**: 465–478. 1946.
12. MILLER, J. J. Studies on the Fusarium of muskmelon wilt. I. Pathogenic and cultural studies with particular reference to the cause and nature of variation in the causal organism. Can. Jour. Res. (C) **23**: 16–43. 1945.
13. PICADO, C. Sur l'action a distance des champignons phytopathogenes. Cong. Path. Veg. Strasbourg., 28–34 (1923). Strasbourg.
14. PRATT, C. A. The staling of fungal cultures; general and chemical investigation of staling by *Fusarium.* Ann. Bot. **38**: 563–594, 599–615. 1924.
15. SAVASTANO, G., and H. S. FAWCETT. A study of decay in citrus fruits produced by inoculations with known mixtures of fungi at different constant temperatures. Jour. Agr. Res. [U.S.] **39**: 163–198. 1929.
16. SCHAFFNIT, E., and M. LÜDTKE. Über die Bildung von Toxinen durch verschiedene Pflanzenparasiten. Ber. Deut. Bot. Ges. **50**: 444–463. 1932.
17. STEINBERG, R. A. A study of some factors in the chemical stimulation of the growth of *Aspergillus niger.* Amer. Jour. Bot. **6**: 330–372. 1919.
18. ————. The growth of fungi in synthetic nutrient solutions. Bot. Rev. **5**: 327–350. 1939.
19. WAKSMAN, S. A. Microbial antagonisms and antibiotic substances. The Commonwealth Fund, New York, 1945.

20. ————————, and E. S. HORNING. Distribution of antagonistic fungi in nature and their antibiotic action. Mycologia **35**: 47–65. 1943.
21. ————————, and H. C. REILLY. The agar streak method for assaying antibiotic substances. Ind. Eng. Chem. (Anal. Ed.) **17**: 556–558. 1945.
22. ————————, and A. SCHATZ. Soil enrichment and development of antagonistic microorganisms. Jour. Bact. **51**: 305–316. 1946.
23. WEINDLING, R. Various fungi recently found to be parasitic on Rhizoctonia solani. (Abstr.) Phytopath. **24**: 1141. 1934.
24. WHITE, R. P. Studies on tomato wilt caused by *Fusarium lycopersici* Sacc. Jour. Agr. Res. [U.S.] **34**: 197–239. 1927.
25. WILKINS, W. H., and G. C. M. HARRIS. Investigation into the production of bacteriostatic substances by fungi. I. Preliminary examination of 100 fungal species. Brit. Jour. Exp. Path. **23**: 166–169. 1942.
26. WOOD, F. C. Studies on "damping off" of cultivated mushrooms and its association with *Fusarium* species. Phytopath. **27**: 85–94. 1937; **29**: 728–739. 1939.

PHYTOPATHOLOGICAL NOTES

Seed Treatment with Mercury Dusts Injurious to Corn with Mechanical Injuries Near Embryo.—Severe injury following seed treatment of corn with mercury dusts applied at normally safe dosages was observed in a field experiment at Madison, Wisconsin, in 1946. Semesan Jr. (1 per cent ethyl mercury phosphate), and Du Pont 1451-D (1.25 per cent ethyl mercury p-toluene sulfonanilide), the two mercury disinfectants included in the experiment with mechanically injured seed, both caused typical mercury injury to nearly all germinating seedlings. The kernels, which had been injured by rupturing the pericarp with a scalpel along the edges of the embryo, were treated on April 13 and planted in the field on May 28. The treated seeds were kept in paper packets in the laboratory, at room temperatures, during the 46-day storage period.

Seeds similarly injured but planted the day they were dusted (April 13) resulted in no mercury damage and responded favorably to the seed treatments with large increases in stands of healthy seedlings over those of the untreated controls. Likewise, seed that had been subjected to crown and tip-cap injuries showed no evidence of mercury injury in either the April or May plantings.

Proof that the mercury injury resulted from storage of the treated seed for an appreciable period before planting, rather than from differences in soil environment when the April and May plantings were made, was established in a subsequent greenhouse experiment. Seed from a mixture of several commercial hybrids was subjected to pericarp injuries as described above and then divided into two lots. One lot was treated with Semesan Jr. immediately after the kernels were injured, and the other 76 days later when both lots were planted in a greenhouse bench, in twelve 25-kernel replications. Nearly every seedling from the seed stored for 76 days before planting developed the stunted and swollen condition typical of mercury injury in contrast with normal seedlings from the seed treated at the time it was planted (Fig. 1). The time of storage required before mercury injury develops in seed having pericarp injuries near the embryo obviously was not determined in these experiments.

The information obtained probably is of no great practical importance but it discloses a fault of mercury dusts not mentioned heretofore in the literature on corn seed treatment.—P. E. Hoppe, Division of Cereal Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, U. S. Department of Agriculture, and the Wisconsin Agricultural Experiment Station.

Sclerotium delphinii Welch on Scilla.[1]—A disease of scilla (*Scilla sibirica*) was observed early in May, 1940, in a commercial planting located

[1] Published as Scientific Paper No. 718, College of Agriculture and Agricultural Experiment Stations, Institute of Agricultural Sciences, State College of Washington, Pullman.

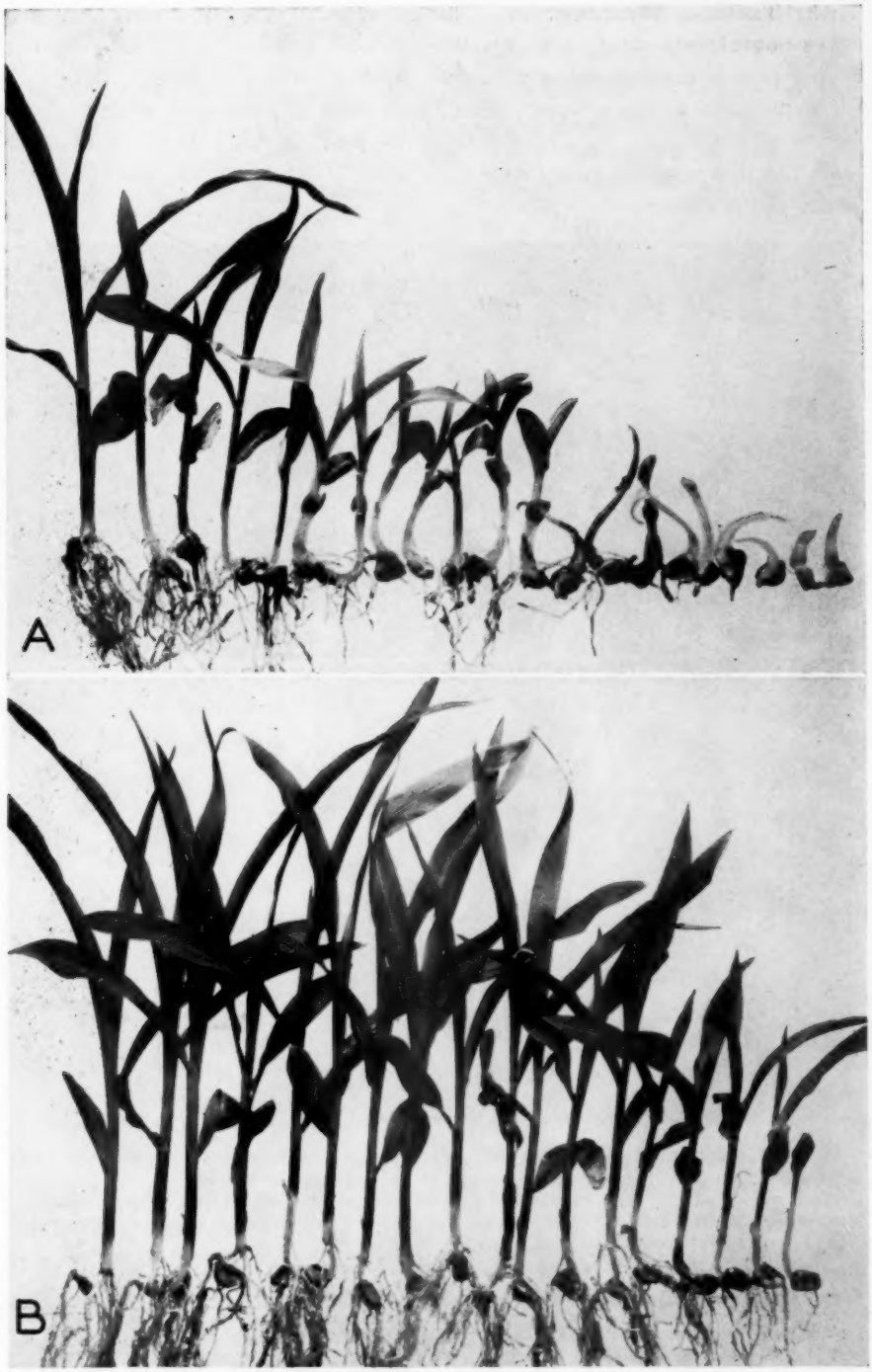

FIG. 1. A. Corn seedlings showing the range of mercury injury (stunting and swollen tissues) resulting from Semesan Jr. treatment of wounded seed stored 76 days before planting; B. Normal seedlings from the same lot of wounded seed planted the day it was dusted.

near Tacoma, Washington. The greater part of the planting (Fig. 1, a) was completely destroyed by the disease, and upon lifting the bulbs, over 90 per cent of the remaining plants were found to be infected.

Symptoms on the above-ground parts were (1) the yellowing and drying up of the leaves, beginning at the tips, and (2) wilting of the blossoms, with the flower stalks remaining erect until the fungus had advanced well above the ground-level.

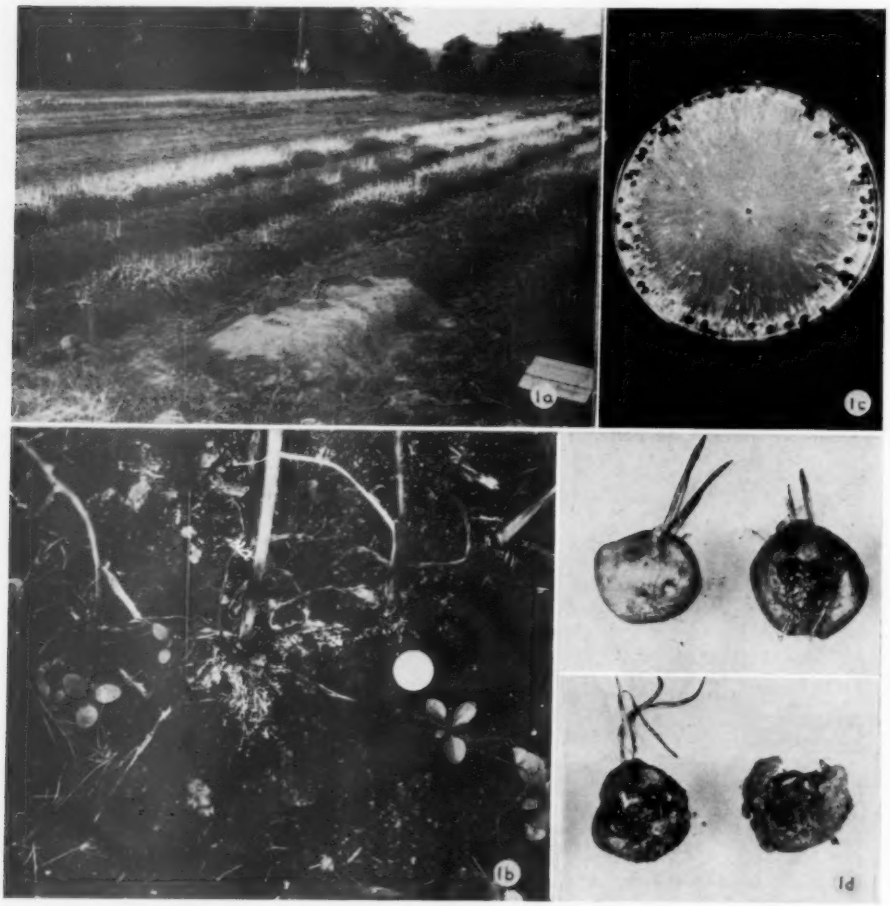

FIG. 1. a. Scilla planting showing destruction of plants by *Sclerotium delphinii*. b. Diseased plant showing mycelial strands on the surface of the soil. c. Plate culture of *Sclerotium delphinii*. d. Scilla bulbs showing various degrees of decay caused by *Sclerotium delphinii*.

Infection apparently started on the bulbs with the fungus advancing from them into the above-ground parts of the plant. Compact strands of mycelium spread over the surface of the soil from the decayed leaves and flower stalks (Fig. 1, b). In the badly infected areas of the beds, fragments of decayed leaves, covered with compact mats of mycelial growth, were

present on the surface of the soil. Reddish-brown to dark brown sclerotia were scattered on the surface and in the soil in the area immediately surrounding the diseased plants.

Infections on the bulbs ranged from small, slightly depressed darkened areas to complete decay. Small dark reddish-brown sclerotia were found in the decayed tissue of the bulbs.

Platings were made on potato-dextrose agar from bulbs showing various stages of infection, from the mycelial mats and strands on the surface of the soil, from sclerotia in the soil, and from infected leaves and flower stalks. White nonsporulating mycelial colonies producing reddish-brown sclerotia in 10 to 14 days (Fig. 1, c) developed from the plantings.

A giant oat-culture of the fungus was mixed with sterilized silt soil in a greenhouse bench and 24 apparently healthy bulbs were planted 6 inches apart in the inoculated soil. Plants from 6 bulbs emerged from the soil, only one of which survived to produce a flower stalk. The remaining 18 bulbs decayed before plant emergence. The same fungus was recovered from all of the infected bulbs. Bulbs planted in non-inoculated sterilized soil developed normally, free from the disease.

One-hundred apparently healthy bulbs were planted in inoculated soil in the field. Four plants failed to emerge from the soil. Examination of these bulbs showed that the bulbs started to decay before the root systems became established. Twenty-two per cent of the plants reached full bloom. Examination of the bulbs five months later revealed lesions varying from 1/16 of an inch in diameter to complete decay of the bulb (Fig. 1, d). Five bulbs showed no apparent infection.

Cultures and partially decayed bulbs were sent to Dr. Donald M. Coe, formerly of the Department of Plant Pathology, State College of Washington, Pullman, Washington, who identified the causal organism as *Sclerotium delphinii* Welch. He stated that the sclerotia of the causal fungus varied only slightly as to size and color from the cultures isolated from Delphinium. However, "it is possible that this may be an additional strain of the fungus."[2]—GLENN A. HUBER and C. J. GOULD, Western Washington Experiment Station, Puyallup, Washington.

Antagonistic Activity of a Species of Actinomyces against Ceratostomella ulmi in vitro.[1]—Increasing knowledge of antibiotics during recent years and recognition of their proved therapeutic value in combating human diseases have stimulated interest in the use of these products for control of plant diseases. Lack of adequate preventive or curative measures for the Dutch elm disease warrants investigation of such materials. This report describes

[2] Quotation from correspondence with Dr. Coe.

[1] Paper of the Journal Series, New Jersey Agricultural Experiment Station, Rutgers University, Department of Plant Pathology.

The author wishes to acknowledge the assistance of the Bartlett Tree Expert Company, Stamford, Conn., who helped finance this work, and of Dr. P. P. Pirone, under whose guidance the work was done.

the antagonistic effect of a species of *Actinomyces* on *Ceratostomella ulmi*, the causal fungus of the disease.

Working with a strain of *Ceratostomella ulmi*, Waksman and Bugie[2] reported in 1943 that the antibiotics actinomycin and clavacin exhibited high activity in laboratory tests against this fungus.

Preliminary tests have indicated that the species of *Actinomyces* used in the present work is distinctly different from *A. antibioticus*, the source of the antibiotic actinomycin. The isolate was originally obtained from C. M. Haenseler who observed it as a contaminant on agar plates.

The isolate of *Ceratostomella ulmi* used in these tests is known as a

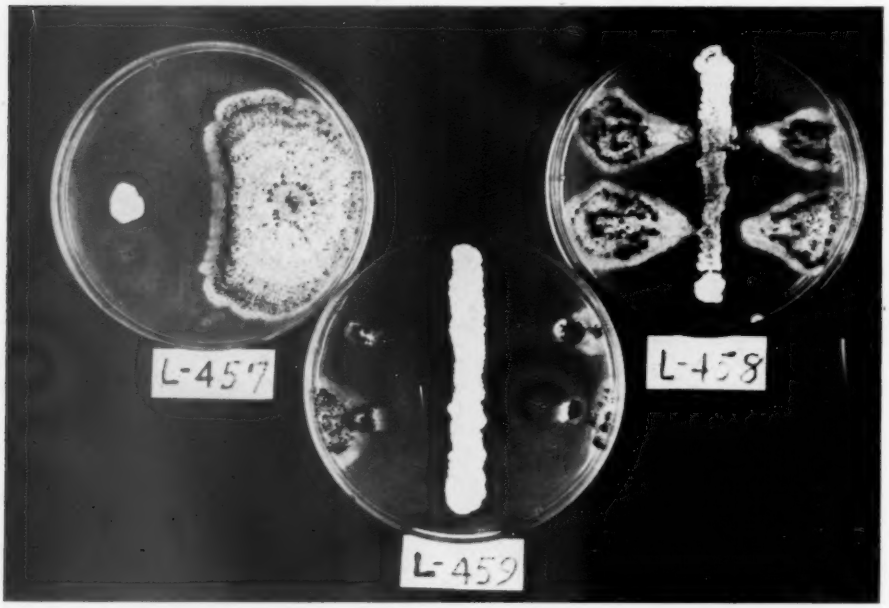

FIG. 1. Antagonistic effect of a species of *Actinomyces* on *Ceratostomella ulmi*. Plates L–457 and L–458: both organisms streaked the same day. Plate L–459: *C. ulmi* streaked 3 days after the *Actinomyces* streak.

"black line strain" and was supplied by the Department of Plant Pathology, Cornell University, under the label, Tyler No. T114a 2aR. It is one of the most virulent strains of the Dutch elm disease fungus thus far isolated from infected trees. Its virulence was tested on 10 healthy trees in May, 1946 before the *in vitro* tests were started. Typical symptoms of the disease appeared on all trees in 8 to 14 days. The organism was recovered in all cases at some distance from the point of inoculation.

Two *in vitro* methods, the agar streak[3] and the two-point inoculation, were used to test the antagonistic action of the *Actinomyces sp.* to *Ceratostomella ulmi*.

[2] Waksman, S. A., and E. Bugie. Action of antibiotic substances upon *Ceratostomella ulmi*. Proc. Soc. Exp. Biol. and Med. 54: 79–82. 1943.

[3] Waksman, S. A., and H. C. Reilly. Agar-streak method for assaying antibiotic substances. Ind. and Eng. Chem. 17: 556. 1945.

In the first method, a long streak of the *Actinomyces sp.* was made across the diameter of a Petri plate containing potato-dextrose agar. Four short streaks (two on each side and at right angles to the long streak) were made with the *Ceratostomella ulmi.* In some plates all streaks were made on the same day; in others, the *C. ulmi* was streaked 1 to 3 days after the *Actinomyces.*

The second method involved the planting of each organism at opposite sides of a poured plate to allow the organisms to grow toward each other. An advantage of this method is that both organisms can be started simultaneously yet sufficient time is provided for the *Actinomyces sp.* to produce its antagonistic substance.

All plates were incubated at 23° to 25° C. and examined periodically.

When the streaks of both organisms were made on the same day, distinct inhibition of *Ceratostomella ulmi* by the *Actinomyces sp.* was evident 1 to 2 days following its streaking. *C. ulmi* did not grow within 20 to 24 mm. of the *Actinomyces* (Fig. 1, L–459).

When the two organisms were planted at opposite sides of the plate, the first indication of the antagonism was observed within 6 days (Fig. 1, L–457). That the inhibitory action began early is indicated by the flattening of the *Ceratostomella ulmi* growth rings, which normally are circular. The zone of inhibition between the *Actinomyces sp.* and *C. ulmi* measured more than 20 mm., and in some cases more than 25 mm. The strong inhibitory activity against *C. ulmi* was retained for 2 months, by which time the medium had dried completely.

The results herein reported both for the streak method and the two-point inoculation method have been repeated more than 10 times with the same consistent results. The antagonistic activity of the *Actinomyces sp.* is entirely independent of pigment formation by the organism.

Efforts to extract the active material from cultures of the *Actinomyces sp.* grown on solid, semi-solid, and liquid media have not succeeded to date. Occasional filtrates and crude extracts with ether show some activity against *Ceratostomella ulmi,* as measured by the cup method technique.—MICHAEL SZKOLNIK, Department of Plant Pathology, Rutgers University, New Brunswick, New Jersey.

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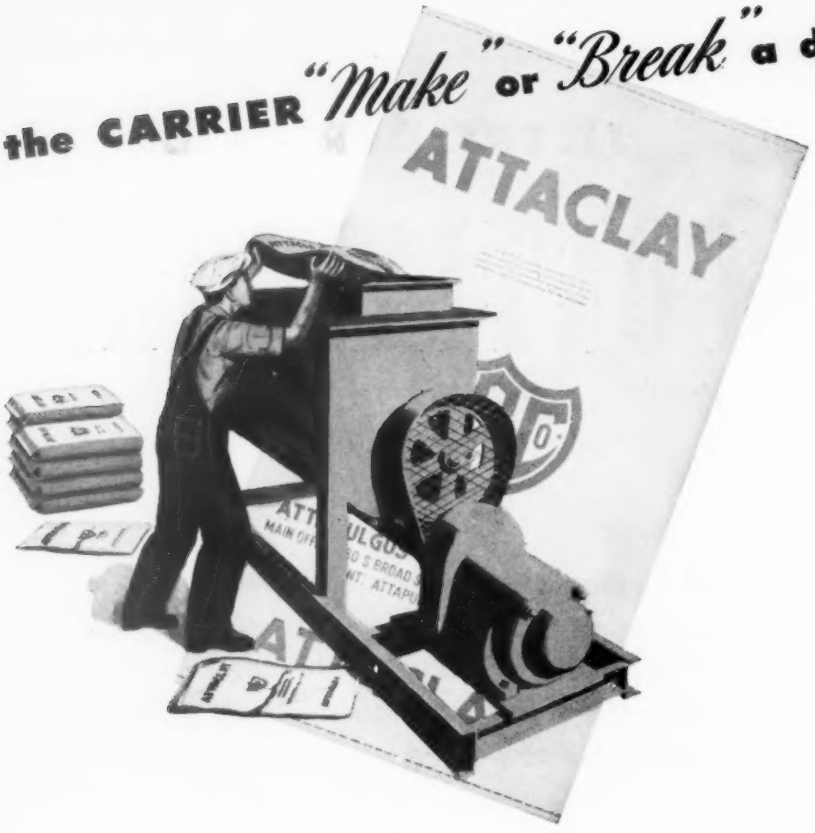

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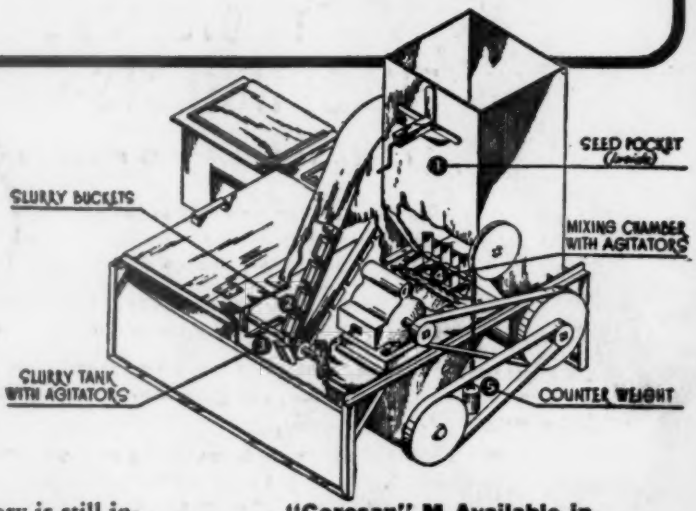

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